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Immunotechnology • Protein Chemistry • High-Performance Dialysis
Molecular Biology • GC Derivatization and Labware

EVERYTHING IS POSSIBLE... WITH THE RIGHT TOOLS®

 **PIERCE**
800-675-5335 815-968-0747

An Introduction to the Pierce Web Site

Page VI

1. New Products

Page NP-1

Featuring Biotinylation and Cross-linking Reagents; Protein L Products, including Immobilized Protein L and Protein L Coated 96-well Plates; SuperSignal® Western and ELISA Chemiluminescent Substrates;

Fluorogenic Peroxidase Substrates; the Glycoprotein Carbohydrate Estimation Kit; RNA and DNA Biotin Labeling Kits; B-PER™ Reagents and Kits; and SnakeSkin™ Dialysis Tubing.

2. Immunotechnology

Section

Page 1

Featuring Carrier Proteins, including Keyhole Limpet Hemocyanin, Ovalbumin, EDC and Bovine Serum Albumin; MagnaBind™ Beads and Supports; Avidin-Biotin Products; Biotinylation Reagents; Complete Antibody Production and Purification Kits; Protein L, Protein A, Protein G and Protein A/G Products; Polyclonal and Monoclonal Antibodies; Pre-coated Microwell Plates; Blocking Buffers; Chemiluminescent, Chromogenic and Fluorogenic Substrates; and Signal Transduction Products.

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3. Protein Chemistry

Section

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Featuring Fluoraldehyde™ *o*-Phthalaldehyde Reagent Solution; Amino Acid Analysis Products; BCA and Coomassie® Assay Reagents and Kits; Cross-linking Reagents, including new Sulfo-SMCC and Sulfo-SBED; BupH™ Dry Buffers; Fluorescent Labels and Probes; IODO-GEN® Pre-Coated Iodination Tubes; Trifluoroacetic Acid; SDS-Out™ Sodium Dodecyl Sulfate Precipitation Reagent; Detergents; GelCode® Blue Stain Reagent; and GelCode® Silver Stain Kit.

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5. Molecular Biology

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Featuring B-PER™ and B-PER™ II Bacterial Protein Extraction Reagents; B-PER™ Column and Spin Purification Kits; B-PER™ Reagent (in Phosphate Buffer); SuperSignal® West HisProbe™ Kit; Immobilized Streptavidin, Avidin and NeutrAvidin™ Products; Biotinylation Reagents; DNA Coating Solution; DAPER Dye DNA Precipitation Agent; and Chemical and Specialty Reagents.

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Featuring Acylation and Silylation Reagents; Column Conditioners; Solvents; Derivatization Reagents; Detergent Concentrates; Reacti-Therm™ Dry Block Sample Derivatization Modules and Accessories; Vials and Closures; Glassware and Accessories; and the Easy-Titer® ELIFA System.

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800-874-3723

NeutrAvidin™ Products

For ultralow nonspecific binding compared to avidin or streptavidin!

Achieve better assay results with the low nonspecific binding properties of NeutrAvidin™. NeutrAvidin™ is a deglycosylated form of avidin, so lectin binding is reduced to undetectable levels without losing biotin-binding affinity ($K_a=10^{15} \text{ M}^{-1}$).¹ NeutrAvidin™ offers the advantage of a neutral pI to minimize nonspecific absorption, along with lysine residues that remain available for derivatization or conjugation through amine-reactive chemistries. The molecular weight of NeutrAvidin™ is approximately 60,000. The specific activity for biotin-binding is approximately 14 µg/mg of protein, which is near the theoretical maximum activity.

Comparison of Avidin, Streptavidin and NeutrAvidin™ Biotin-Binding Protein

	Isoelectric Point	Contains Carbohydrate	Contains Recognition Sequence RYD	Degree of Nonspecific Binding
Avidin	10.0	Yes	No	High
Streptavidin	5.0	No	Yes	Low
NeutrAvidin™	6.3	No	No	Lowest

Reference

1. Hiller, Y., Gershoni, J.M., Bayer, E.A. and Wilchek, M. (1987). Biotin binding to avidin. Oligosaccharide side chain not required for ligand association. *Biochem. J.* 248, 167-171.

Ordering Information

Product #	Description	Features/Benefits	Pkg. Size	U.S. Price
31000ZZ	ImmunoPure® NeutrAvidin™ Biotin-Binding Protein	<ul style="list-style-type: none"> pI that has been reduced to a neutral state Deglycosylated, so lectin binding is reduced to undetectable levels Can use as a biotin blocking agent in tissues for histochemistry 	10 mg	\$ 91
31001ZZ	NeutrAvidin™, Horseradish Peroxidase Conjugated	<ul style="list-style-type: none"> Lower nonspecific binding than streptavidin conjugates Better signal-to-noise ratio in assay systems 	2 mg	\$150
31002ZZ	NeutrAvidin™, Alkaline Phosphatase Conjugated	<ul style="list-style-type: none"> Lower nonspecific binding than streptavidin conjugates Better signal-to-noise ratio in assay systems 	2 mg	\$185
31006ZZ	NeutrAvidin™, Fluorescein Conjugated	<ul style="list-style-type: none"> Fluorescent-labeled NeutrAvidin™ Biotin-Binding Protein Absorption: 490 nm; Emission 520 nm 	5 mg	\$115
31005ZZ	NeutrAvidin™, Goat Anti-Mouse IgG Conjugated	<ul style="list-style-type: none"> Can achieve a 5- to 8-fold amplification in your immunoassay Better signal-to-noise ratio than avidin or streptavidin 	1 mg	\$ 89
31007ZZ	EZ-Link™ Maleimide Activated NeutrAvidin™ Biotin-Binding Protein	<ul style="list-style-type: none"> Pre-activated for ease of use Reacts spontaneously with free thiols in the pH range of 6.5-7.5 	5 mg	\$ 75

Purified Streptavidin and Conjugates

Lower nonspecific binding than avidin.

Pierce offers streptavidin products that produce high-quality results. The molecular weight of streptavidin is about 60,000. Unlike avidin, streptavidin has no carbohydrate and has an acidic isoelectric point of 5. Streptavidin is much less soluble in water than avidin and can be crystallized from water or 50% isopropanol. Streptavidin is also a tetrameric protein, and each subunit binds one molecule of biotin. Guanidine hydrochloride will dissociate avidin and streptavidin into subunits, but streptavidin is more resistant to dissociation. ImmunoPure® Streptavidin products are affinity-purified and have a specific activity greater than 12 units/mg.

Features/Benefits:

- Isolated from *Streptomyces avidinii* and purified over an affinity column to assure the integrity of the four biotin-binding sites
- Lower noise level (less nonspecific binding) than avidin in detection systems

Applications for enzyme-labeled streptavidin:

- Enzyme immunoassays (EIA)
- Cell and tissue staining (for light microscopy)
- Blot immunostaining

Applications for fluorochrome-labeled streptavidin:

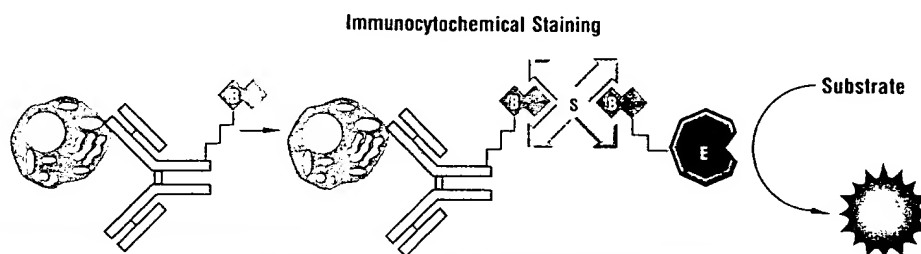
- Immunofluorescence assays (IFA)
- Cell staining (for fluorescent microscopy)
- Tissue staining
- Blot immunostaining
- Cell staining (for fluorescent-activated cell sorting)

Note: 1 unit of biotin binding equals 1 µmole of biotin.

Reference

1. Chalet, I. and Wolf, F.J. (1964). The properties of streptavidin, a biotin-binding protein produced by *Streptomyces*. *Arch. Biochem. Biophys.* 106, 1-5.

Purified Streptavidin and Conjugates *continued*



Ordering Information

Product #	Description	Features/Benefits	Applications	Pkg. Size	U.S. Price
21122ZZ	ImmunoPure™ Streptavidin	<ul style="list-style-type: none"> • Lyophilized, stable powder • Molecular mass of 60,000 daltons • No carbohydrate and has an acidic isoelectric point of 5 • Much less soluble in water than avidin • Specific activity greater than 12 units of biotin binding/mg of protein 	<ul style="list-style-type: none"> • Immunoassay reagent when bound to biotinylated enzymes or when conjugated to enzymes • Blocking protein for biotin-rich tissue sections (use at 0.1% for inhibition of endogenous biotin) 	1 mg	\$109
21125ZZ	ImmunoPure™ Streptavidin	<ul style="list-style-type: none"> • 1-2 moles of HRP to streptavidin • 80-150 units of HRP activity per mg of conjugate • Lyophilized 	<ul style="list-style-type: none"> • Histochemistry • Western blotting 	5 mg	\$235
21126ZZ	Streptavidin, Horseradish Peroxidase Conjugated	<ul style="list-style-type: none"> • 2-5 units of biotin-binding activity per mg of conjugate • 300-400 units of AP activity per mg of conjugate 	<ul style="list-style-type: none"> • Histochemistry • Western blotting 	1 mg	\$105
21124ZZ	Streptavidin, Horseradish Peroxidase Conjugated	<ul style="list-style-type: none"> • Fluorescent-labeled streptavidin • Excitation: 490 nm • Emission: 520 nm • Color: Greenish-yellow 	<ul style="list-style-type: none"> • Histochemical staining • Fluorescent-activated cell sorting 	2 mg	\$199
21127ZZ	Streptavidin, Horseradish Peroxidase Conjugated	<ul style="list-style-type: none"> • Fluorescent-labeled streptavidin • Absorption: 515-520 nm and 550-555 nm • Emission: 575 nm • Color: Orange-red 	<ul style="list-style-type: none"> • Histochemical staining; can be used in double staining methods • Fluorescent-activated cell sorting 	5 mg	\$345
21324ZZ	Streptavidin, Alkaline Phosphatase Conjugated	<ul style="list-style-type: none"> • Fluorescent-labeled streptavidin • Absorption: 595 nm • Emission: 615 nm • Color: Orange-red 	<ul style="list-style-type: none"> • Used to create immunoassay reagents • Localize glycoproteins on blot transfers, followed by detection with a biotinylated enzyme 	1 mg	\$139
21323ZZ	Streptavidin, Alkaline Phosphatase Conjugated	<ul style="list-style-type: none"> • Attaches streptavidin to oxidized carbohydrate residues on glycoproteins 	<ul style="list-style-type: none"> • Histochemical staining • Fluorescent-activated cell sorting 	3 mg	\$320
21224ZZ	Streptavidin, Fluorescein (FITC) Conjugated	<ul style="list-style-type: none"> • Attaches streptavidin to oxidized carbohydrate residues on glycoproteins 	<ul style="list-style-type: none"> • Histochemical staining • Fluorescent-activated cell sorting 	1 mg	\$136
21724ZZ	Streptavidin, Rhodamine (TRITC) Conjugated	<ul style="list-style-type: none"> • Attaches streptavidin to oxidized carbohydrate residues on glycoproteins 	<ul style="list-style-type: none"> • Histochemical staining • Fluorescent-activated cell sorting 	1 mg	\$133
21624ZZ	Streptavidin, Texas Red™ Conjugated	<ul style="list-style-type: none"> • Attaches streptavidin to oxidized carbohydrate residues on glycoproteins 	<ul style="list-style-type: none"> • Histochemical staining • Fluorescent-activated cell sorting 	1 mg	\$139
21120ZZ	ImmunoPure™ Streptavidin Hydrazide	<ul style="list-style-type: none"> • Attaches streptavidin to oxidized carbohydrate residues on glycoproteins 	<ul style="list-style-type: none"> • Histochemical staining • Fluorescent-activated cell sorting 	2 mg	\$ 90

References

1. Savage, M.D., Mattson, G., Desai, S., Nielander, G.W., Morgensen, S. and Conklin, E.J. (1992). *Avidin-Biotin Chemistry: A Handbook*. Rockford, Illinois: Pierce Chemical Company. (Product # 15055ZZ)
2. Wilchek, M. and Bayer, E.A. (1983). The avidin-biotin complex in bioanalytical applications. *Anal. Biochem.* 71, 1-32.

EZ-Link™ Biotinylating Reagents

Attach biotin to almost any molecule the easy way!


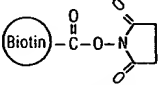
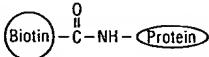

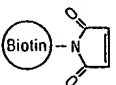
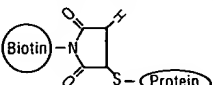

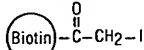
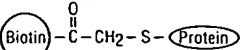
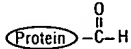
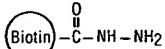
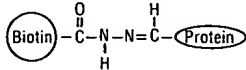
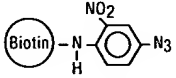

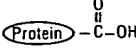
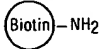
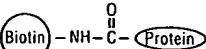
Pierce has created a line of high-purity, biotinylating reagents to help you develop unique assay reagents. Whether you are derivatizing DNA or a glycoprotein, Pierce has a wide selection of EZ-Link™ Biotinylating Reagents to fit your application. Once a compound is biotinylated, it can be used to create a variety of assay systems to detect antigens or DNA/RNA without the use of radioactivity. Attach a biotin group to primary amine-containing compounds with Sulfo-NHS-LC-Biotin (Product # 21335ZZ) and create a stable amide bond that can withstand many assay buffer conditions. Some advantages of directly labeling a primary antibody instead of using a commercially available, biotinylated secondary antibody for immunoassays are:

- Reduced number of steps in the immunoassay
- Reduction in the amount of time to complete the immunoassay

- Reduced amount of background by decreasing the effects of cross-reactivity
- Improved tissue penetration for immunohistochemical applications
- Reduction in the number of washing steps

Reactive chemistries on the biotinylating reagents react with available functional groups on the molecule to be biotinylated. Use the table below as a guide for selecting the right chemistry for your application. Other factors to take into consideration when selecting a biotinylating reagent are:

- Solubility in aqueous solutions or organic solvents
- Length of the spacer arm between the biotin group and the other molecule
- Whether or not the spacer arm is cleavable

Functional Group	Reactive Group	Linkage Formed
Primary Amine 	NHS-Ester/Sulfo-NHS Ester  PRODUCTS: NHS-LC-Biotin, Sulfo-NHS-LC-Biotin, NHS-LC-LC-Biotin, Sulfo-NHS-LC-LC-Biotin, NHS-Biotin, Sulfo-NHS-Biotin, NHS-SS-Biotin	Amide Bond 
Sulfhydryl 	Maleimide  PRODUCTS: Biotin-BMCC	Thioether Bond 
Sulfhydryl 	Iodoacetyl  PRODUCTS: Iodoacetyl-LC-Biotin, PEO-Iodoacetyl Biotin	Thioether Bond 
Oxidized Carbohydrate 	Hydrazide  PRODUCTS: Biotin Hydrazide, Biotin-LC-Hydrazide, Biocytin Hydrazide	Hydrazone Bond 
DNA/RNA Protein Carbohydrates	Azido (Photoactivatable)  PRODUCTS: Photoactivatable Biotin, Biotin-LC-ASA	 Ring expansion followed by coupling with primary amine
Carboxyl  (Reaction requires EDC cross-linker)	Other  PRODUCTS: (Biotinamido)pentylamine	Amide Bond 

Biotinylation Reagent Selection Guide

Product #	Description	Chemical Reactivity	Water-Soluble	Spacer Arm Length	Cleavable	Membrane-Permeable*
21335ZZ	Sulfo-NHS-LC-Biotin	Primary Amine	Yes	22.4 Å	No	No
21338ZZ	Sulfo-NHS-LC-LC-Biotin	Primary Amine	Yes	30.5 Å	No	No
21217ZZ	Sulfo-NHS-Biotin	Primary Amine	Yes	13.5 Å	No	No
21331ZZ	Sulfo-NHS-SS-Biotin	Primary Amine	Yes	24.3 Å	Yes	No
21336ZZ	NHS-LC-Biotin	Primary Amine	No	22.4 Å	No	Yes
21343ZZ	NHS-LC-LC-Biotin	Primary Amine	No	30.5 Å	No	Yes
20217ZZ	NHS-Biotin	Primary Amine	No	13.5 Å	No	Yes
21117ZZ	NHS-Iminobiotin	Primary Amine	No	13.5 Å	No	Yes
21218ZZ	PFP-Biotin	Primary or Secondary Amines/RNA/DNA	No	9.6 Å	No	Yes
21332ZZ	NHS-PC-LC-Biotin	Primary Amine	No	31.4 Å	No	Yes
21901ZZ	PEO-Maleimide Activated Biotin	Sulfhydryl	Yes	29.1 Å	No	No
21900ZZ	Biotin-BMCC	Sulfhydryl	No	32.6 Å	No	Yes
21334ZZ	PEO-Iodoacetyl Biotin	Sulfhydryl	Yes	24.7 Å	No	No
21333ZZ	Iodoacetyl-LC-Biotin	Sulfhydryl	No	27.1 Å	No	Yes
21341ZZ	Biotin-HPDP	Sulfhydryl	No	29.2 Å	Yes	Yes
21346ZZ	Biotin PEO-Amine	Carboxyl	Yes	20.4 Å	No	No
21347ZZ	Biotin PEO-LC-Amine	Carboxyl	Yes	22.9 Å	No	No
21345ZZ	5-(Biotinamido)pentylamine	Carboxyl	Yes	18.9 Å	No	No
28020ZZ	Biocytin Hydrazide	Carbohydrate/RNA/DNA	Yes	19.7 Å	No	No
21339ZZ	Biotin Hydrazide	Carbohydrate	No	15.7 Å	No	Yes
21340ZZ	Biotin-LC-Hydrazide	Carbohydrate	No	24.7 Å	No	Yes
29986ZZ	Psoralen-PEO-Biotin	DNA/RNA/Protein	Yes	36.9 Å	No	No
22020ZZ	PEO-Biotin Dimer	Avidin Cross-linking	Yes	43.4 Å	No	No
29987ZZ	Photoactivatable Biotin	DNA/RNA/Protein	No	30.0 Å	No	Yes
29982ZZ	Biotin-LC-ASA	DNA/RNA/Protein	No	29.9 Å	No	Yes
28023ZZ	p-Aminobenzoyl Biocytin	DNA/RNA/Protein	Yes	27.9 Å	Yes	No
28022ZZ	Biocytin	Hydrazide	Yes	20.1 Å	No	No
22008ZZ	Biotin DPPE	Phospholipids	No	42.0 Å	No	No
22010ZZ	Biotin-LC-DPPE	Phospholipids	No	50.3 Å	No	No
33033ZZ	Sulfo-SBED	Trifunctional	Yes	N/A	Yes	No

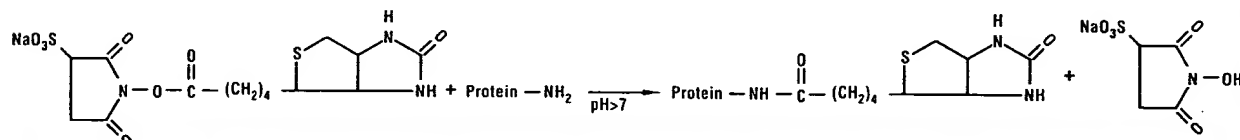
*Membrane permeability is implied due to a molecule's hydrophobic/hydrophilic nature

EZ-Link™ Biotinylation Reagents Reactive Toward Amines

EZ-Link™ Biotinylation Kits

Contain everything you need for convenient, successful biotinylations.

The Reaction of Sulfo-NHS-Biotin (Water Soluble) With a Protein

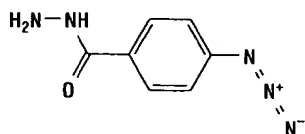


Ordering Information

Product #	Description	Pkg. Size	U.S. Price	Product #	Description	Pkg. Size	U.S. Price
21420ZZ	EZ-Link™ Sulfo-NHS-Biotinylation Kit Includes: EZ-Link™ Sulfo-NHS-Biotin PBS, Lyophilized Sodium Phosphate (0.1 M), Lyophilized Dextran Plastic Desalting Column HABA (10 mM) Avidin, Affinity-purified	Kit 25 mg 1 x 10 ml 1 ml 10 mg	\$185	21430ZZ	EZ-Link™ Sulfo-NHS-LC-Biotinylation Kit Includes: EZ-Link™ Sulfo-NHS-LC-Biotin PBS, Lyophilized Sodium Phosphate (0.1 M), Lyophilized Dextran Plastic Desalting Column HABA (10 mM) Avidin, Affinity-purified	Kit 25 mg 1 x 10 ml 1 ml 10 mg	\$199

ABH

A carbohydrate-reactive, photoactivatable cross-linker.



ABH
M.W. 177.16
Spacer Arm 11.9 Å

Features/Benefits:

- Hydrazide group of ABH reacts with *cis*-diol containing carbohydrates, proteins or other molecules, after the carbohydrates have been oxidized to form aldehydes
- Arylazide end of ABH reacts nonspecifically with proteins or other molecules upon UV photolysis

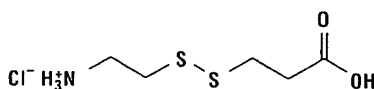
- Reactive groups: hydrazide and phenyl azide
- Reactive toward: oxidized carbohydrate and amino groups
- Literature reference #1 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21510ZZ	ABH (<i>p</i> -Azidobenzoyl Hydrazide)	50 mg	\$178

AEDP

Applications in reversible immobilization and cross-linking.



AEDP
M.W. 217.74
Spacer Arm 9.5 Å

Features/Benefits:

- Cleavable amino acid that can be used as a spacer arm in conjugation schemes
- Reversible immobilization application
- Cleavable by DTT, 2-mercaptoethylamine or TCEP
- Affects incorporation of amine or carboxyl groups into proteins/peptides via use of cross-linking agents

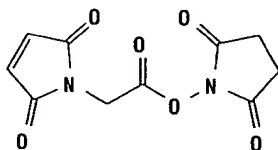
- The amine end of the linker can be reacted with amine-reactive acylation agents, yielding amide bonds
- Use with water-soluble carbodiimide (EDC) to conjugate AEDP with amines or carboxylates on target molecules
- Reactive groups: amine and carboxyl
- Reactive toward: NHS esters/Sulfo-NHS esters and amines/hydrazides via EDC activation
- Literature reference #'s 55, 56 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22101ZZ	AEDP (3-[(2-Aminoethyl)dithio]propionic acid-HCl)	50 mg	\$ 65

AMAS

A short aliphatic spacer for amine-sulphydryl cross-linking.



AMAS
M.W. 252.18
Spacer Arm 4.4 Å

Features/Benefits:

- Non-cleavable, close proximity cross-linker
- Aliphatic spacer has low potential for eliciting an immune response
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds

- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Functional homolog of BMPS, GMBS and EMCS
- Water-insoluble
- Reactive groups: maleimide and NHS ester
- Reactive toward: sulphydryl and amino groups
- Literature reference #'s 57, 58 (page 214)

Ordering Information

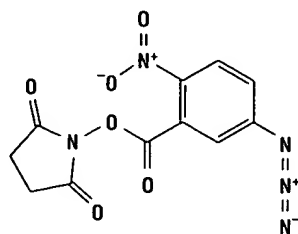
Product #	Description	Pkg. Size	U.S. Price
22295ZZ	AMAS* (<i>N</i> -[α -Maleimidoacetoxy]succinimide ester)	50 mg	\$ 70

*See also: GMBS and EMCS and their sulfonated analogs, Sulfo-GMBS, Sulfo-EMCS, Sulfo-KMUS.

800-874-3723

ANB-NOS

Nitro group on the phenyl azide shifts the optimal wavelength for photolysis to 320-350 nm, which helps to maintain the biological integrity of proteins and nucleic acids.



ANB-NOS
M.W. 305.20
Spacer Arm 7.7 Å

Features/Benefits:

- *Reactive groups:* NHS ester and nitrophenyl azide
- *Reactive toward:* amino groups
- Literature reference #2 (page 213)

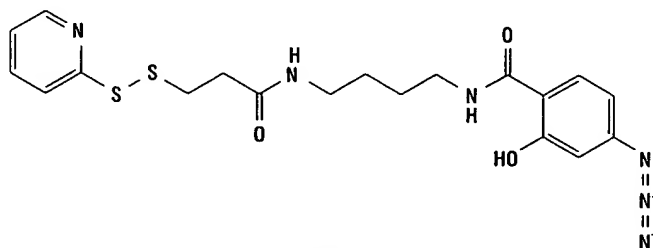
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21451ZZ	ANB-NOS* (N-5-Azido-2-nitrobenzoyloxysuccinimide)	50 mg	\$ 45

*Sulfonated, water-soluble analog also available; see SAND.

APDP

A radioiodinatable, cleavable, sulfhydryl-reactive and photoreactive cross-linker.



APDP
M.W. 446.55
Spacer Arm 21.0 Å

Features/Benefits:

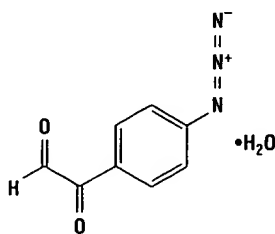
- APDP reacts first with a free sulfhydryl-containing ligand by disulfide exchange between the cross-linker and a free sulfhydryl on the ligand (often from cysteine residues)
- *Reactive groups:* pyridyldisulfide and hydroxyphenyl azide
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #3 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27720ZZ	APDP (N-[4-(p-Azidosalicylamido)butyl]-3'-(2'-pyridyldithio)propionamide)	50 mg	\$199

APG

Reacts selectively with arginine residues at pH 7-8.



APG
M.W. 193.16
Spacer Arm 9.3 Å

Features/Benefits:

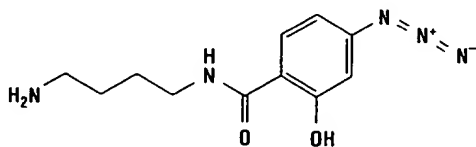
- *Reactive groups:* phenyl azide and phenylglyoxal
- *Reactive toward:* amino groups and guanidium side chain of arginine
- Literature reference #4 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20108ZZ	APG (p-Azidophenyl glyoxal monohydrate)	50 mg	\$ 81

ASBA

A photoreactive, iodlatable and carboxyl-reactive cross-linker.



ASBA
M.W. 249.27
Spacer Arm 16.3 Å

Features/Benefits:

- Amine terminal of ASBA will react with a carboxylic acid in the presence of a dehydrating reagent, such as EDC, to form an amide bond; amide product is then ready for iodination and subsequent photo-coupling

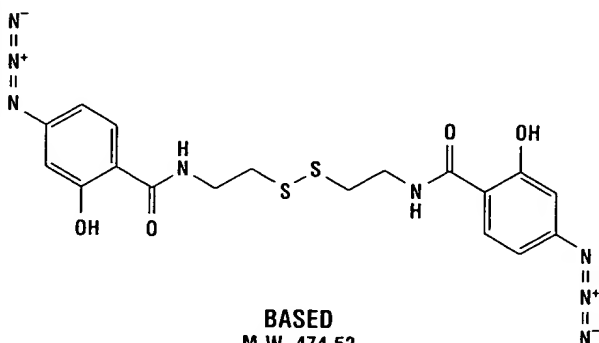
- *Reactive groups:* amine and hydroxy phenyl azide
- *Reactive toward:* carboxyl groups and amines
- Literature reference #5 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21512ZZ	ASBA (4-[p-Azidosalicylamido]butylamine)	50 mg	\$ 95

BASED

Contains two photoreactive phenyl azides for nonspecific conjugation of proteins.



BASED
M.W. 474.52
Spacer Arm 34.7 Å

Features/Benefits:

- Phenyl azides of BASED are iodlatable, so each protein will have a radioactive label after the photolysis reaction

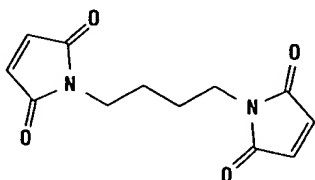
- Because BASED reacts nonspecifically with proteins and other biomolecules, it is useful when a ligand contains neither amines nor sulfhydryls in a location appropriate to obtain the desired conjugate
- Cleavable by reducing agents
- *Reactive groups:* hydroxy phenyl azide (homobifunctional)
- *Reactive toward:* amines
- Literature reference #6 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21564ZZ	BASED (Bis-[β-(4-azidosalicylamido)ethyl]disulfide)	50 mg	\$199

BMB

A non-cleavable, homobifunctional, sulfhydryl-reactive cross-linker with a four carbon spacer.



BMB
M.W. 248.23
Spacer Arm 10.9 Å

Features/Benefits:

- Intermediate length cross-linker
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages

- Non-cleavable analog of BMDB
- Water-insoluble
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups
- Literature reference #'s 7, 59-62 (pages 213-214)

Ordering Information

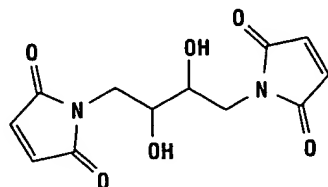
Product #	Description	Pkg. Size	U.S. Price
22331ZZ	BMB* (1,4-Bis-maleimidobutane)	50 mg	\$ 65

*See also: BMDB, BMOE, BMH, DTME, HBVS.

800-874-3723

BMDB

A sulfhydryl-reactive homobifunctional cross-linker cleaved by periodate.



BMDB
M.W. 280.23
Spacer Arm 10.2 Å

Features/Benefits:

- Bis-maleimido, vic-diol-containing agent
- Cross-links sulfhydryl-containing compounds under mild conditions

- Specific reactivity with -SH groups at pH 6.5-7.5
- Cleavage with sodium periodate preserves indigenous S-S bonds and tertiary structure
- Cross-links reversed by treatment with 15 mM sodium periodate
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups
- Literature reference #'s 59-62 (page 214)

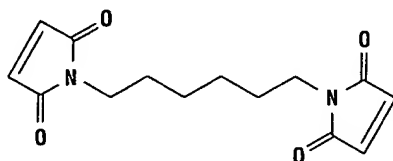
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22332ZZ	BMDB* (1,4 Bis-Maleimidyl-2,3-dihydroxybutane)	50 mg	\$ 69

*European Patent # 9710209

BMH

Mild irreversible sulfhydryl-to-sulfhydryl cross-linking agent.



BMH
M.W. 276.29
Spacer Arm 16.1 Å

Features/Benefits:

- BMH permits irreversible cross-linking of sulfhydryl-containing compounds, using mild conditions
- Maleimides react specifically with sulfhydryl groups at pH 6.5-7.5

- Water-insoluble; non-cleavable
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups
- Literature reference #7 (page 213)

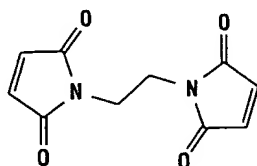
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22330ZZ	BMH* (Bis-Maleimido-hexane)	50 mg	\$ 54

*See also: BMOE, BMB, BMDB, DTME, HBVS.

BMOE

Short spacer sulfhydryl-to-sulfhydryl cross-linking.



BMOE
M.W. 220.18
Spacer Arm 8.0 Å

Features/Benefits:

- Shortest bis-maleimide cross-linker available for close proximity cross-linking
- Non-cleavable; water-insoluble

- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- C₂ homolog of BMH
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups
- Literature reference #7 (page 213)

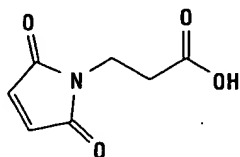
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22323ZZ	BMOE* (Bis-Maleimidoethane)	50 mg	\$ 65

*See also: BMH, BMB, BMDB.

BMPA

Modifies sulphydryl groups to carboxyl groups for the preparation of peptide-protein conjugates.



BMPA
M.W. 169.13
Spacer Arm 5.9 Å

Features/Benefits:

- Sulphydryl modification agent that creates a terminal -COOH group at -SH sites in proteins and other molecules
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages

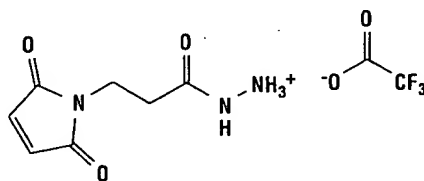
- Makes -COOH groups available for coupling via EDC activation
- Non-cleavable
- Use as protein modification agent or cross-linker
- *Reactive groups:* maleimide and carboxyl
- *Reactive toward:* sulphydryl groups and amines/hydrazides
- Literature reference #'s 5, 63, 64 (pages 213-214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22296ZZ	BMPA (N-β-Maleimidopropionic acid)	100 mg	\$ 29

BMPH

Water-soluble, heterobifunctional cross-linker facilitates glycoconjugate formation.



BMPH
M.W. 297.19
Spacer Arm 8.1 Å

Features/Benefits:

- Sulphydryl-reactive and carbonyl-reactive heterobifunctional reagent
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages

- Water-soluble; non-cleavable
- Useful for covalently coupling to oxidized carbohydrate moieties in glycoproteins and other glycoconjugates
- *Reactive groups:* maleimide and hydrazide
- *Reactive toward:* sulphydryl groups and carbonyl (aldehyde)/carboxyl groups
- Literature reference #84 (page 214)

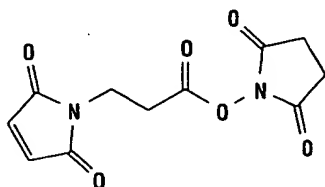
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22297ZZ	BMPH* (N-β-Maleimidopropionic acid) hydrazide•TFA)	50 mg	\$ 72

*See also: M₂C₂H, MPBH, PDPH.

BMPS

Short spacer amine-to-sulphydryl cross-linking agent for the preparation of immunoconjugates.



BMPS
M.W. 266.21
Spacer Arm 6.9 Å

Features/Benefits:

- C₃ homolog of aliphatic spacer series (AMAS, GMBS and EMCS) with identical reactivities
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds
- Non-cleavable; water-insoluble

- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Aliphatic spacer has low potential for eliciting an immune response, ensuring that the primary response to the antigen-carrier protein is not diluted by a response against a determinant on the cross-linker
- *Reactive groups:* maleimide and NHS ester
- *Reactive toward:* sulphydryl and amine groups
- Literature reference #'s 32, 43 (pages 213-214)

Ordering Information

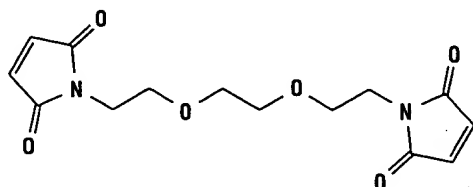
Product #	Description	Pkg. Size	U.S. Price
22298ZZ	BMPS* (N-β-Maleimidopropylsuccinimidyl ester)	50 mg	\$ 70

*See also: AMAS, GMBS, EMCS.

800-874-3723

BM[PEO]₃

Eight-atom polyether spacer reduces potential for conjugate precipitation in sulfhydryl-to-sulfhydryl cross-linking applications.



BM[PEO]₃
M.W. 308.29
Spacer Arm 14.7 Å

Features/Benefits:

- Long sulfhydryl-reactive, homobifunctional cross-linker
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages

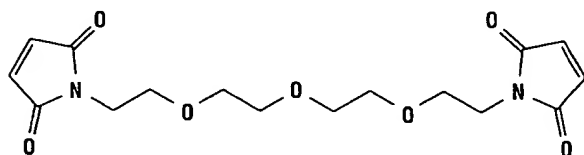
- Non-cleavable; water-soluble
- Polyethylene oxide (PEO) cross-bridge provides increased water solubility, reducing the potential for cross-linker-caused precipitation of conjugates
- Ideal for small molecule or peptide conjugations
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22336ZZ	BM[PEO] ₃ (1,8-Bis-Maleimidotriethyleneglycol)	50 mg	\$ 39

BM[PEO]₄

Eleven atom polyether spacer provides more reach and reduces potential for conjugate precipitation.



BM[PEO]₄
M.W. 352.34
Spacer Arm 17.8 Å

Features/Benefits:

- Long sulfhydryl-reactive, homobifunctional cross-linker
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Non-cleavable; water-soluble

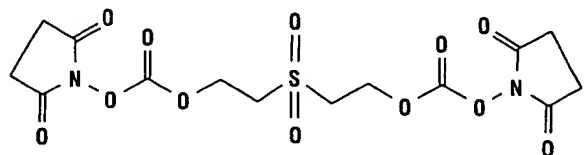
- Polyethylene oxide (PEO) cross-bridge provides increased water solubility, reducing the potential for cross-linker-caused precipitation of conjugates
- Ideal for small molecule or peptide conjugations
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22337ZZ	BM[PEO] ₄ (1,11-Bis-Maleimidotetraethyleneglycol)	50 mg	\$ 71

BSOCOES

Base-reversible cross-linking reagent.



BSOCOES
M.W. 436.35
Spacer Arm 13.0 Å

Features/Benefits:

- Water-insoluble
- Base-cleavable (pH 11.6, 2 hours, 37°C)

- *Reactive groups:* NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #'s 8, 39 (page 213)

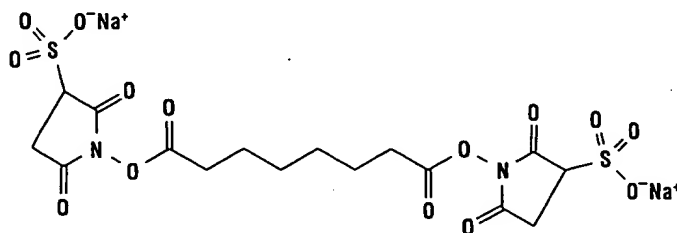
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21600ZZ	BSOCOES* (Bis[2-(Succinimidyl)oxycarbonyloxy]-ethyl)sulfone)	50 mg	\$ 59

*Sulfonated, water-soluble analog also available; see Sulfo-BSOCOES.

BS³

A popular non-cleavable, membrane-impermeable, water-soluble DSS analog.



BS³
M.W. 572.43
Spacer Arm 11.4 Å

Features/Benefits:

- Water-soluble DSS analog
- Non-cleavable
- Membrane-impermeable
- *Reactive groups:* Sulfo-NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #9 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21580ZZ	BS ³ (Bis [Sulfosuccinimidyl]suberate)	50 mg	\$ 54

DFDNB

Useful for cross-linking between small spatial distances.



DFDNB
M.W. 204.09
Spacer Arm 3.0 Å

Features/Benefits:

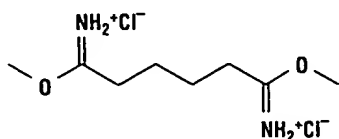
- *Reactive groups:* aryl halide
- *Reactive toward:* amino groups
- Literature reference #10 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21525ZZ	DFDNB (1,5-Difluoro-2,4-dinitrobenzene)	1 gm	\$ 49

DMA

Cross-links outer membrane proteins.



DMA
M.W. 245.15
Spacer Arm 8.6 Å

Features/Benefits:

- Rapid reaction with amines at alkaline pH values (pH 8-10)
- Amidine bond retains net charge character of protein

- Reversible at high pH values
- Tool for study of quaternary structure of proteins
- *Reactive groups:* imidoester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #11 (page 213)

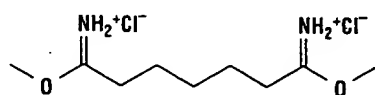
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20663ZZ	DMA (Dimethyl adipimidate•2 HCl)	50 mg	\$ 25

800-874-3723

DMP

Covalently links an oriented antibody to Protein A.



DMP
M.W. 259.17
Spacer Arm 9.2 Å

Features/Benefits:

- Rapid reaction with amines at alkaline pH values (pH 8-10)
- Amidine bond retains net charge character of protein
- Reversible at high pH values
- Tool for study of quaternary structure of proteins

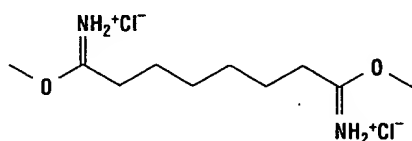
- *Reactive groups:* imidoester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #12 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21666ZZ	DMP (Dimethyl pimelimidate•2 HCl)	50 mg	\$ 20

DMS

A longer chain length imidoester cross-linker.



DMS
M.W. 273.20
Spacer Arm 11.0 Å

Features/Benefits:

- Rapid reaction with amines at alkaline pH values (pH 8-10)
- Amidine bond retains net charge character of protein
- Reversible at high pH values
- Tool for study of quaternary structure of proteins

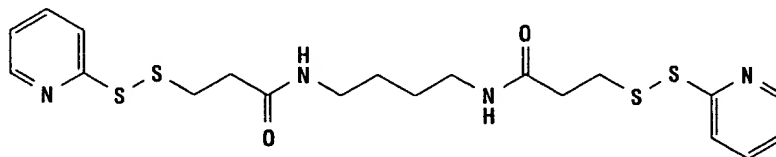
- *Reactive groups:* imidoester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #13 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20700ZZ	DMS (Dimethyl suberimidate•2 HCl)	1 gm	\$ 30

DPDPB

A cleavable, sulfhydryl-reactive homobifunctional cross-linker.



DPDPB
M.W. 482.71
Spacer Arm 19.9 Å

Features/Benefits:

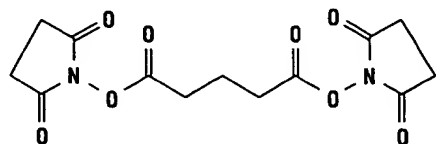
- Forms a mixed disulfide with sulfhydryl-containing compounds; disulfide can easily be cleaved with an appropriate reducing agent
- *Reactive groups:* pyridyldithio
- *Reactive toward:* sulfhydryl groups
- Literature reference #14 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21702ZZ	DPDPB (1,4-Di-[3'-(2'-pyridyldithio)-propionamido]butane)	50 mg	\$ 71

DSG

Homobifunctional, non-cleavable five carbon spacer homolog of DSS.



DSG
M.W. 326.26
Spacer Arm 7.7 Å

Features/Benefits:

- Can increase the cross-linking efficiency compared to that of DSS in some applications

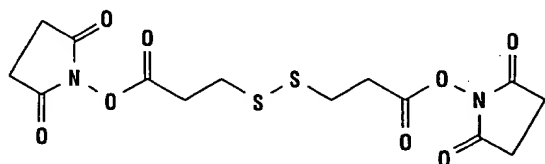
- *Reactive groups:* NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #15 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20593ZZ	DSG (Disuccinimidyl glutarate)	50 mg	\$ 35

DSP (Lomant's Reagent)

Thiol-cleavable, homobifunctional and amine-reactive.



DSP
M.W. 404.42
Spacer Arm 12.0 Å

Features/Benefits:

- Acylate in aqueous or organic media within two minutes at room temperature
- Four- to five-hour half-life of ester at pH 7.0
- Introduced disulfides quantitatively cleaved at 37°C with 10-50 mM DTT at pH 8.5 within 30 minutes

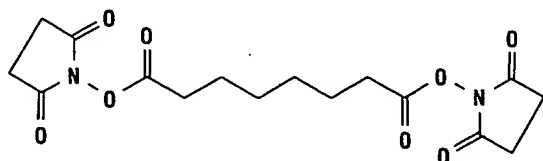
- Disulfides also cleaved with 5% 2-mercaptoethanol in SDS-PAGE sample buffer (2% SDS, 0.25 mM Tris base, 10% glycerol) at 100°C for 5 minutes
- *Reactive groups:* NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #16 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22585ZZ	DSP (Lomant's Reagent) (Dithiobis(succinimidyl propionate))	1 gm	\$112

DSS

A non-cleavable, amine-reactive, homobifunctional cross-linker suited to receptor-protein studies.



DSS
M.W. 368.35
Spacer Arm 11.4 Å

Features/Benefits:

- Used for conjugating a radiolabeled ligand to a cell surface receptor
- Water-insoluble; non-cleavable

- *Reactive groups:* NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #17 (page 213)

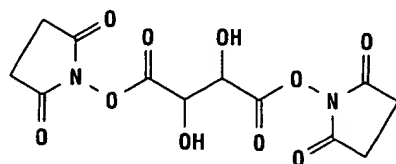
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21555ZZ	DSS (Disuccinimidyl suberate)	1 gm	\$ 87

• 800-874-3723

DST

Unique cross-linker cleavable by oxidizing reagents.



DST
M.W. 344.24
Spacer Arm 6.4 Å

Features/Benefits:

- Ideal for applications in which cross-link reversibility is desired without disturbing protein S-S bonds

- *Reactive groups:* NHS esters (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #'s 18, 40 (page 213)

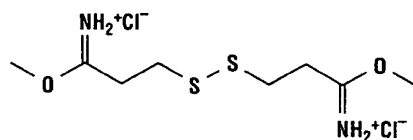
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20589ZZ	DST* (Disuccinimidyl tartrate)	50 mg	\$ 45

*Sulfonated, water-soluble analog also available; see Sulfo-DST.
See also: BMDB.

DTBP

A cleavable, bifunctional, imidoester cross-linker.



DTBP
M.W. 309.28
Spacer Arm 11.9 Å

Features/Benefits:

- Penetrates intact human erythrocytes
- See DMA for imidoester characteristics

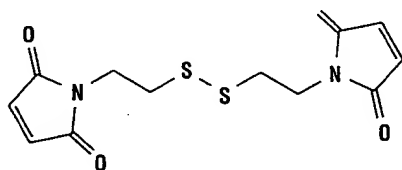
- *Reactive groups:* imidoesters (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #19 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20665ZZ	DTBP (Wang and Richards Reagent) (Dimethyl 3,3'-dithiobispropionimidate•2 HCl)	1 gm	\$ 93

DTME

Cleavable sulfhydryl-to-sulfhydryl cross-linking agent.



DTME
M.W. 312.37
Spacer Arm 13.3 Å

Features/Benefits:

- Intermediate length, sulfhydryl-reactive cross-linker
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages

- Cleavable by DTT, 2-mercaptoethanol or TCEP
- Water-insoluble
- Cleavable analog of BMH
- *Reactive groups:* maleimide (homobifunctional)
- *Reactive toward:* sulfhydryl groups
- Literature reference #'s 7, 65 (pages 213-214)

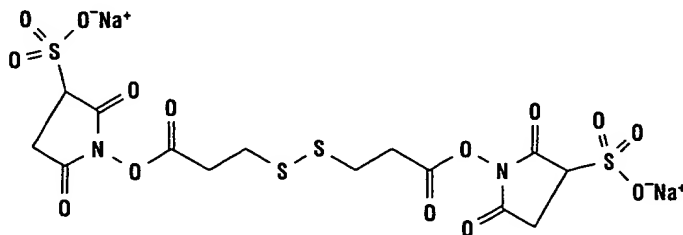
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22335ZZ	DTME* (Dithio-bis-maleimidoethane)	50 mg	\$159

*See also: BMH, BMDB, DPDBP.

DTSSP

A water-soluble, membrane-impermeable, thiol-cleavable cross-linker.



DTSSP
M.W. 608.51
Spacer Arm 12.0 Å

Features/Benefits:

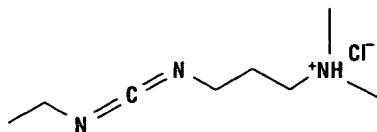
- Conditions for reducing the disulfide bond include: 50 mM DTT, 100 mM β-mercaptoethanol or 1% sodium borohydride
- *Reactive groups:* Sulfo-NHS esters (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #20 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21578ZZ	DTSSP (3,3'-Dithiobis[sulfosuccinimidyl propionate])	50 mg	\$ 59

EDC

A water-soluble carbodiimide for rapid preparation of peptide conjugates.



EDC
M.W. 191.70

Features/Benefits:

- Easy removal of excess reagent and corresponding urea after coupling by washing with dilute acid or water
- Numerous chemical conjugates can be synthesized via available -COOH and -NH₂ groups
- Amide bond formed provides a neutral linkage, which is ideal for preparing peptides and antigens

- Addition of Sulfo-NHS enhances the coupling reaction at physiological pH values
- *Reactive group:* carbodiimide
- *Reactive toward:* amino groups
- Literature reference #'s 21, 88 (pages 213-214)

Ordering Information

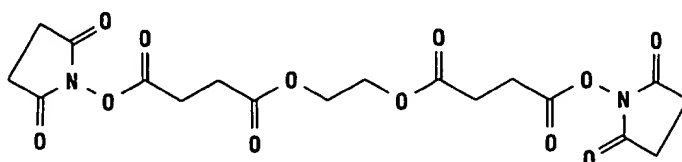
Product #	Description	Pkg. Size	U.S. Price
22980ZZ	EDC* (1-Ethyl-3-[3-dimethylaminopropyl]carbodiimide Hydrochloride)	5 gm	\$ 61
22981ZZ	EDC	25 gm	\$198

*See also Sulfo-NHS (Product # 24510ZZ).

800-874-3723

EGS

Long reach and cleavable under gentle conditions.



EGS
M.W. 456.36
Spacer Arm 16.1 Å

Features/Benefits:

- Cross-links formed are cleavable at pH 8.5 using hydroxylamine (Product # 26103ZZ) for three to six hours at 37°C
- Lactose dehydrogenase retained 60% of its activity after reversible cross-linking with EGS
- *Reactive groups:* NHS esters (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #'s 22, 41 (page 213)

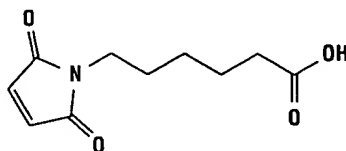
Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
21565ZZ	EGS* (Ethylene glycol bis[succinimidylsuccinate])	1 gm	\$ 98

*Sulfonated, water-soluble analog also available; see Sulfo-EGS.

EMCA

Useful for introducing maleimide groups into biomolecules.



EMCA
M.W. 211.21
Spacer Arm 9.4 Å

Features/Benefits:

- Sulfhydryl-reactive and amine-reactive via water-soluble carbodiimide (EDC) coupling
- Prepare maleimide-activated proteins through EDC coupling of the carboxyl group to available protein amino groups

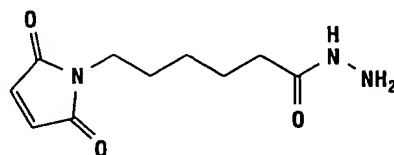
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Modifies protein sulfhydryl groups to carboxyl groups
- Non-cleavable
- Probe for protein -SH groups
- *Reactive groups:* maleimide and carboxyl
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #66 (page 214)

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
22306ZZ	EMCA (N-ε-Maleimidocaproic acid)	1 gm	\$109

EMCH

Sulfhydryl- and carbohydrate-reactive cross-linker with immune response neutral cross-bridge.



EMCH
M.W. 225.24
Spacer Arm 11.8 Å

Features/Benefits:

- Sulfhydryl- and oxidized carbohydrate-reactive agent
- Covalently couple the hydrazide group of EMCH to glycoproteins and other glycoconjugates after oxidation with sodium periodate

- Maleimide reacts with -SH groups at a pH of 6.5-7.5, forming stable thioether linkages
- *Reactive groups:* maleimide and hydrazide
- *Reactive toward:* sulfhydryl and carbonyl (oxidized carbohydrate) groups
- Literature reference #67 (page 214)

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
22106ZZ	EMCH* (N-ε-Maleimidocaproic acid)hydrazide)	50 mg	\$107

*See also: M₂C₄H, MPBH, PDPH.

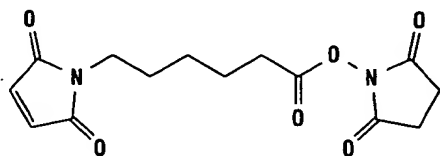
3

Internet: <http://www.sigma-aldrich.com>

184

EMCS

Amine-sulphydryl-reactive linker with low immunogenicity and a little more reach.



EMCS
M.W. 308.29
Spacer Arm 9.4 Å

Features/Benefits:

- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages

- Non-cleavable; water-insoluble
- Increased sphere of coupling vs. GMBS
- Aliphatic spacer offers low potential for eliciting an immune response
- *Reactive groups:* maleimide and NHS ester
- *Reactive toward:* sulphydryl and amino groups
- Literature reference #'s 23, 68, 69 (pages 213-214)

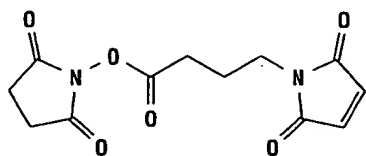
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22308ZZ	EMCS* (N-[ε-Maleimidocaproyloxy]succinimide ester)	50 mg	\$ 70

*Sulfonated, water-soluble analog also available; see Sulfo-EMCS.

GMBS

Heterobifunctional analog of MBS – enhanced maleimide stability with less immunogenicity.



GMBS
M.W. 280.23
Spacer Arm 6.8 Å

Features/Benefits:

- Non-cleavable
- Low potential for eliciting an immune response, ensuring that the primary response to the antigen-carrier protein conjugate is not diluted by a response against a determinant on the cross-linker

- Less immunogenic than SMCC
- *Reactive groups:* NHS ester and maleimide
- *Reactive toward:* amino and sulphydryl groups
- Literature reference #'s 23, 84 (pages 213-214)

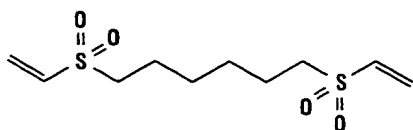
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22309ZZ	GMBS* (N-[γ-Maleimidobutyryloxy]succinimide ester)	50 mg	\$ 70

*Sulfonated, water-soluble analog also available; see Sulfo-GMBS.

HBVS

Sulphydryl reactivity without the hydrolysis potential of maleimides.



HBVS
M.W. 266.38
Spacer Arm 14.7 Å

Features/Benefits:

- Novel sulphydryl-reactive, homobifunctional cross-linking agent
- Couples via Michael addition, yielding stable thioether links without stereoisomer formation

- Vinylsulfone-reactive group is indefinitely stable at pH 7 and resists hydrolytic degradation for days at pH 9
- Non-cleavable; water-insoluble
- *Reactive group:* vinylsulfone (homobifunctional)
- *Reactive toward:* sulphydryl groups
- Literature reference #'s 78-80 (page 214)

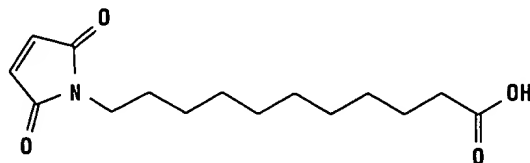
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22334ZZ	HBVS (1,6-Hexane-bis-vinylsulfone)	50 mg	\$ 71

• 800-874-3723

KMUA

Activate biomolecules for cross-linking through sulfhydryl groups or introduce carboxyl groups into proteins.



KMUA
M.W. 281.35
Spacer Arm 15.7 Å

Features/Benefits:

- Novel sulfhydryl-reactive, heterobifunctional cross-linking agent
- Maleimide activate protein/peptide via EDC activation of the carboxyl group
- Sulfhydryl modification agent that creates a terminal carboxylate group at -SH sites in proteins and other molecules

- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Non-cleavable, long aliphatic cross-bridge
- Useful for preparing peptide-protein conjugates
- *Reactive groups:* maleimide and carboxyl
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 63, 64 (page 214)

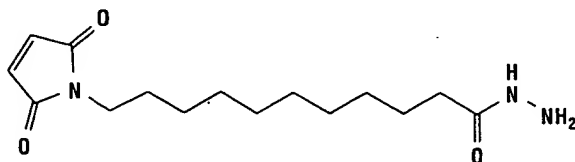
Ordering Information

Product #	Description	Pkg Size	U.S. Price
22211ZZ	KMUA* (N- κ -Maleimidoundecanoic acid)	100 mg	\$ 29

*See also: BMPA, EMCA.

KMUH

Extended chain length; heterobifunctional for the preparation of glycoconjugates.



KMUH
M.W. 295.38
Spacer Arm 19.0 Å

Features/Benefits:

- Sulfhydryl-reactive and carbonyl-reactive heterobifunctional cross-linking agent
- Long, non-cleavable, aliphatic cross-bridge

- Hydrazide group covalently couples to oxidized carbohydrate residues in glycoproteins and other glycoconjugates
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- *Reactive groups:* maleimide and hydrazide
- *Reactive toward:* sulfhydryl and carbonyl (aldehyde)/carboxyl groups
- Literature reference #67 (page 214)

Ordering Information

Product #	Description	Pkg Size	U.S. Price
22111ZZ	KMUH* (N- κ -Maleimidoundecanoic acid)hydrazide)	50 mg	\$121

*See also: BMPH, EMCH, M₂C₂H, MPBH, PDPH.

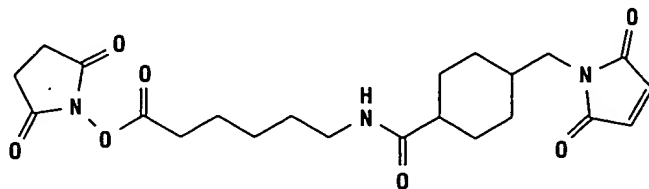
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Internet: <http://www.merck.com>

3-3

LC-SMCC

Extended chain length analog of SMCC, a popular reagent for immunoconjugate preparation.



LC-SMCC
M.W. 447.48
Spacer Arm 16.1 Å

Features/Benefits:

- Sulfhydryl-reactive and amine-reactive heterobifunctional cross-linking agent
- SMCC and analogs are ideal for coupling enzymes to antibodies as both enzyme activity and antibody specificity can be preserved after coupling
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Cyclohexane containing cross-bridge stabilizes the maleimide group

- Non-cleavable; water-insoluble
- Useful for the preparation of stable maleimide activated proteins
- *Reactive groups:* NHS ester and maleimide
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 35, 52, 70 (pages 213-214)

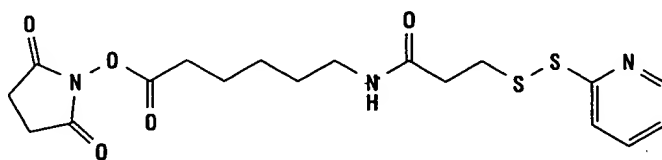
Ordering Information

Product #	Description	Pkg Size	U.S. Price
22362ZZ	LC-SMCC* (Succinimidyl-4-[N-maleimidomethyl]-cyclohexane-1-carboxy-[6-amidocaproate])	50 mg	\$ 89

*See also: SMCC, Sulfo-SMCC.

LC-SPDP

Classic heterobifunctional, cleavable cross-linker, with an extended spacer arm.



LC-SPDP
M.W. 425.52
Spacer Arm 15.7 Å

Features/Benefits:

- LC-SPDP releases a detectable byproduct after reacting with a free sulfhydryl group; by measuring the release of pyridine-2-thione at 343 nm, the reaction can be easily followed
- *Reactive groups:* pyridyldithio and NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 33, 38 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21651ZZ	LC-SPDP* (Succinimidyl 6-[3-(2-pyridyldithio)-propionamido]hexanoate)	50 mg	\$245

*Sulfonated, water-soluble analog also available; see Sulfo-LC-SPDP.

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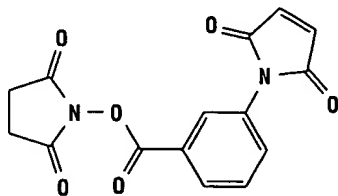
MOBIUM CHEMISTRY

3-3

800-874-3723

MBS

Useful for coupling proteins, enzymes to antibodies, toxins to antibodies, and haptens to carrier proteins.



MBS
M.W. 314.25
Spacer Arm 9.9 Å

Features/Benefits:

- Water-insoluble
- Non-cleavable

- Popular for forming enzyme immunoconjugates
- *Reactive groups:* NHS ester and maleimide
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 25, 42, 84 (pages 213-214)

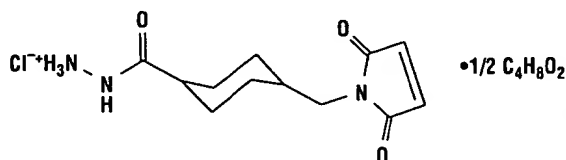
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22311ZZ	MBS* (<i>m</i> -Maleimidobenzoyl- <i>N</i> -hydroxysuccinimide ester)	50 mg	\$ 41

*Sulfonated, water-soluble analog also available; see Sulfo-MBS.
See also: SMPB.

M₂C₂H

Combine carbohydrate selectivity with sulfhydryl reactivity.



M₂C₂H
M.W. 331.8
Spacer Arm 15.1 Å

Features/Benefits:

- Has an oxidized carbohydrate-specific hydrazide, a sulfhydryl-reactive group and spacer arm to accommodate a wide range of molecular coupling demands
- Sulfhydryl-specific group is a maleimide that yields a thioether linkage upon coupling
- Stable in acetonitrile

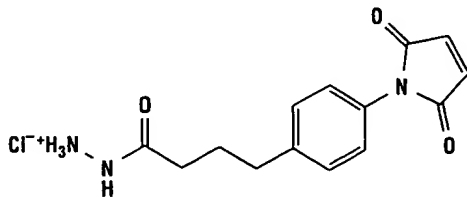
- *Reactive groups:* hydrazide and maleimide
- *Reactive toward:* carbohydrate and sulfhydryl groups
- Literature reference #26 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22303ZZ	M ₂ C ₂ H (4-[(<i>N</i> -Maleimidomethyl)cyclohexane-1-carboxylhydrazide]·HCl·1/2 dioxane)	50 mg	\$10

MPBH

Carbohydrate-selective and sulfhydryl-reactive.



MPBH
M.W. 309.75
Spacer Arm 17.9 Å

Features/Benefits:

- Has an oxidized carbohydrate-specific hydrazide, a sulfhydryl-reactive group and spacer arm to accommodate a wide range of molecular coupling demands

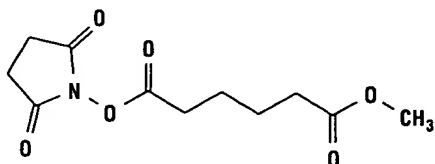
- Sulfhydryl-specific group is a maleimide that yields a thioether linkage upon coupling
- Stable in DMSO
- *Reactive groups:* hydrazide and maleimide
- *Reactive toward:* carbohydrate and sulfhydryl groups
- Literature reference #26 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22305ZZ	MPBH (4-[(<i>N</i> -Maleimidophenyl)butyric acid hydrazide]·HCl)	50 mg	\$10

MSA

Amine-reactive with latent carboxyl group.



MSA
M.W. 257.24
Spacer Arm 7.2 Å

Features/Benefits:

- Amine-reactive modification reagent containing a masked carboxyl group
- NHS ester reacts with primary amines at pH 7-9 to form stable amide bonds

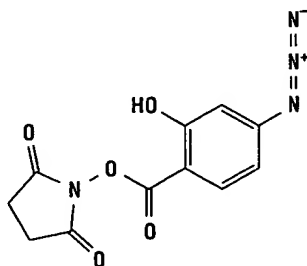
- Masked carboxyl group containing heterobifunctional reagent
- Converts amino groups to carboxyl groups
- Carboxyl group freed at pH 9.5 in phosphate buffer
- Non-cleavable; water-insoluble
- *Reactive groups:* NHS ester and methyl ester
- *Reactive toward:* amino groups

Ordering Information

Product #	Description	Pkg Size	U.S. Price
22605ZZ	MSA (Methyl <i>N</i> -succinimidyl adipate)	50 mg	\$ 45

NHS-ASA

An ¹²⁵I label can be easily and effectively incorporated into this reagent before the acylation step, and used to radiolabel conjugates.



NHS-ASA
M.W. 276.21
Spacer Arm 8.0 Å

Features/Benefits:

- Photolysis reaction is readily initiated by long wave UV light
- NHS-ASA has been used to simplify detection of photo-affinity labeled complexes and the determination of cross-linked loci
- *Reactive groups:* hydroxyphenyl azide and NHS ester
- *Reactive toward:* amino groups
- Literature reference #27 (page 213)

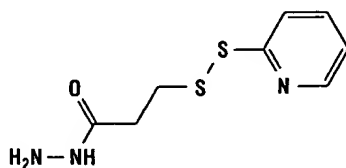
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27714ZZ	NHS-ASA* (<i>N</i> -Hydroxysuccinimidyl-4-azidosalicylic acid)	50 mg	\$ 65

*Sulfonated, water-soluble analogs also available; see Sulfo-NHS-ASA and Sulfo-LC-NHS-ASA.

PDPH

A cleavable, carbohydrate-selective, sulfhydryl-reactive cross-linker.



PDPH
M.W. 229.32
Spacer Arm 9.2 Å

Features/Benefits:

- Pyridyl disulfide group of PDPH gives a cleavable disulfide linkage in the conjugate

- An oxidized carbohydrate-specific hydrazide
- *Reactive groups:* pyridyldithio and hydrazide
- *Reactive toward:* sulfhydryl groups and carbohydrate
- Literature reference #'s 28, 76, 77 (pages 213-214)

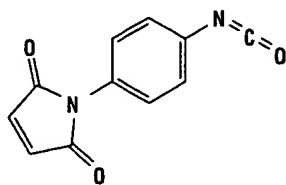
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22301ZZ	PDPH (3-[2-Pyridyldithio]propionyl hydrazide)	50 mg	\$102

• 800-874-3723

PMPI

Both hydroxyl and sulfhydryl reactivity can be found in this unique cross-linker.



PMPI
M.W. 214.18
Spacer Arm 8.7 Å

Features/Benefits:

- Novel sulfhydryl- and hydroxyl-reactive heterobifunctional cross-linker
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages

- Isocyanate reacts with -OH groups to form a carbamate link at pH 8.5
- Non-cleavable
- Excellent tool for the preparation of conjugates of -OH group containing compounds such as steroids and vitamins
- *Reactive groups:* maleimide and isocyanate
- *Reactive toward:* sulfhydryl and hydroxyl groups
- Literature reference #71 (page 214)

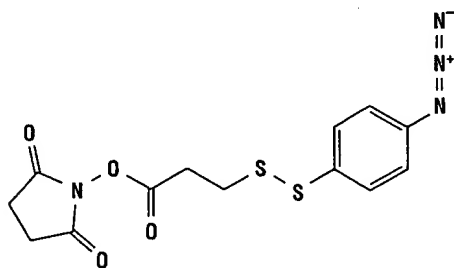
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28100ZZ	PMPI* (N-[p-Maleimidophenyl]isocyanate)	50 mg	\$ 86

*See also: BMPA, EMCA.

SADP

Versatile, cleavable and photoreactive cross-linker.



SADP
M.W. 352.39
Spacer Arm 13.9 Å

Features/Benefits:

- Cleavable by 50 mM dithiothreitol, 100 mM β -mercaptoethanol or 1% sodium borohydride
- Photolysis is achieved by irradiation at 265-275 nm

- *Reactive groups:* phenylazide and NHS ester
- *Reactive toward:* amino groups
- Literature reference #53 (page 214)

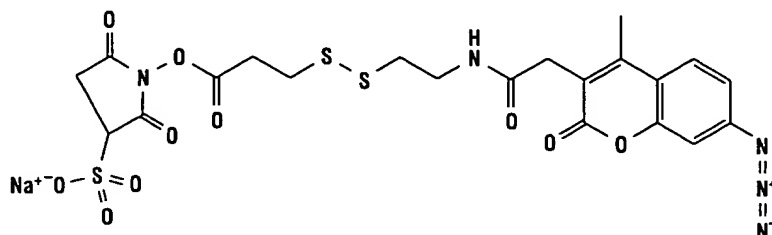
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21533ZZ	SADP* (N-Succinimidyl [4-azidophenyl]-1,3'-dithiopropionate)	50 mg	\$ 73

*Sulfonated, water-soluble analog also available; see Sulfo-SADP.

SAED

Cleavable and photoreactive – a safer, easier alternative to radiolabeling proteins.



SAED
M.W. 621.60
Spacer Arm 23.6 Å

Features/Benefits:

- N-sulfosuccinimidyl ester terminal of SAED will react with amino groups of one protein; the phenyl azide terminal can be reacted nonspecifically under ultraviolet conditions to link a second protein
- Disulfide bond of SAED may be cleaved with an appropriate reducing agent
- *Reactive groups:* azido-methylcoumarin and Sulfo-NHS ester

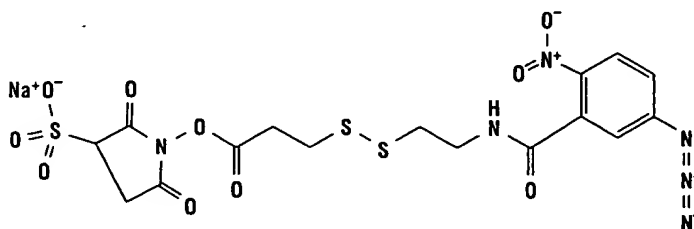
- *Reactive toward:* amino groups
- Literature reference #54 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
33030ZZ	SAED (Sulfosuccinimidyl 2-[7-azido-4-methylcoumarin-3-acetamido]ethyl-1,3'-dithiopropionate)	5 mg	\$103

SAND

Water-soluble, long chain, cleavable analog of ANB-NOS.



SAND
M.W. 570.51
Spacer Arm 18.5 Å

Features/Benefits:

- Cleavable by thiols
- Nitro group on the phenyl azide allows for photolysis at 320-350 nm
- *Reactive groups:* Sulfo-NHS ester and nitrophenyl azide
- *Reactive toward:* amino groups
- Literature reference #29 (page 213)

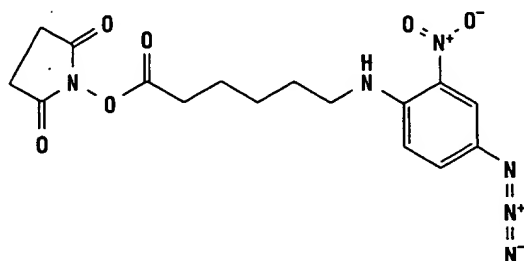
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21549ZZ	SAND (Sulfosuccinimidyl 2-[m-azido-o-nitrobenzamido]ethyl-1,3'-dithiopropionate)	50 mg	\$131

800-874-3723

SANPAH

Extended chain length, photoactivatable cross-linker.



SANPAH
M.W. 390.35
Spacer Arm 18.2 Å

Features/Benefits:

- Optimal photolysis occurs at 320-350 nm; a condition that limits damage to biomolecules by irradiation

- *Reactive groups:* nitrophenyl azide and NHS ester
- *Reactive toward:* amino groups
- Literature reference #30 (page 213)

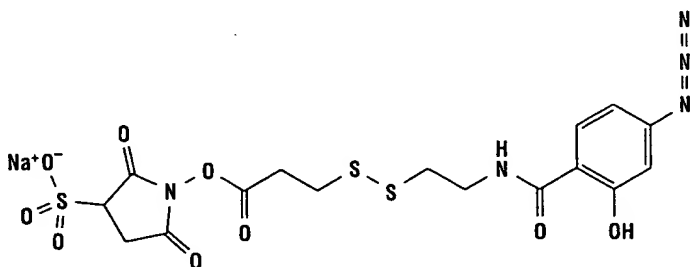
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22600ZZ	SANPAH* (N-Succinimidyl 6-[4'-azido-2'-nitro-phenylamino]hexanoate)	50 mg	\$ 69

*Sulfonated, water-soluble analog also available; see Sulfo-SANPAH.

SASD

Transfers a radioactive label from one protein to another.



SASD
M.W. 541.51
Spacer Arm 18.9 Å

Features/Benefits:

- Radioiodinatable,* cleavable, photoreactive, heterobifunctional cross-linker
- Iodination occurs between the azide and the hydroxyl groups of the phenyl ring; after cleavage by a reducing agent, the radioactive label will remain attached to the protein conjugated by photolysis

- *Reactive groups:* hydroxyphenyl azide and Sulfo-NHS ester
- *Reactive toward:* amino groups
- Literature reference #'s 31, 51 (pages 213-214)

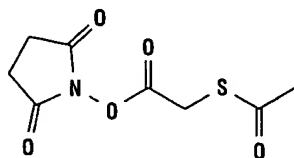
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27716ZZ	SASD (Sulfosuccinimidyl-2-[p-azido-salicylamido]ethyl-1,3'-dithiopropionate)	50 mg	\$16

See IODO-GEN Iodination Reagent (Product #'s 28600ZZ and 28601ZZ).

SATA

Adds protected sulfhydryls for better control when a sulfhydryl group is needed in conjugate formation.



SATA
M.W. 231.23
Spacer Arm 2.8 Å

Features/Benefits:

- Reacts with amines to add protected sulfhydryl groups
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds
- Converts amino groups to sulfhydryl groups
- Latent -SH group is released by hydroxylamine and is available for reaction with maleimide-activated biomolecules or other -SH group-containing compounds

- Cross-links formed with other -SH group-containing molecules are reversible by reducing agents
- *Reactive groups:* NHS ester and thioacetyl-protected sulfhydryl
- *Reactive toward:* amino and maleimide/iodoacetyl or vinyl sulfone
- Literature reference #72 (page 214)

Ordering Information:

Product #	Description	Pkg Size	U.S. Price
26102ZZ	SATA* (N-Succinimidyl S-acetylthioacetate)	50 mg	\$ 30

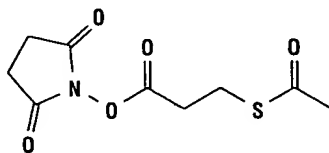
*See also: SATP.

Compatible Pierce Products for Use With SATA and SATP

20688ZZ	DMSO	950 ml	\$ 29
26103ZZ	Hydroxylamine•HCl	25 gm	\$ 28

SATP

Same function as SATA, but offers more steric freedom for the unmasked sulfhydryl group.



SATP
M.W. 245.25
Spacer Arm 4.1 Å

Features/Benefits:

- Thiolation reagent that reacts with amines to add protected sulfhydryl groups
- Converts amino groups to sulfhydryl groups
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds
- Latent -SH group is released by hydroxylamine treatment and available for reaction with maleimide-activated biomolecules or other -SH group-containing compounds

- Cross-links formed with other -SH group-containing molecules are reversible by reducing agents
- Modify peptides to facilitate the preparation of hapten-carrier conjugates
- *Reactive groups:* NHS ester and thioacetyl-protected sulfhydryl
- *Reactive toward:* amino and maleimide/iodoacetyl or vinyl sulfone
- Literature reference #'s 72, 81 (page 214)

Ordering Information:

Product #	Description	Pkg Size	U.S. Price
26100ZZ	SATP (N-Succinimidyl S-acetylthiopropionate)	50 mg	\$ 49

Compatible Pierce Products for Use With SATP

20688ZZ	DMSO	950 ml	\$ 29
26103ZZ	Hydroxylamine•HCl	25 gm	\$ 28

3

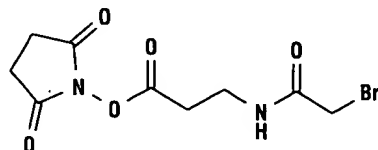
Protein Chemistry

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• 800-874-3723

SBAP

Alternative active halogen chemistry applied to the preparation of cyclic peptides and peptide conjugates.



SBAP
M.W. 307.10
Spacer Arm 6.2 Å

Features/Benefits:

- NHS ester reacts with primary amines at pH 7-9 to form a stable amide bond
- Bromoacetyl group reacts with sulfhydryl groups at pH > 7.5 to form stable thioether bonds

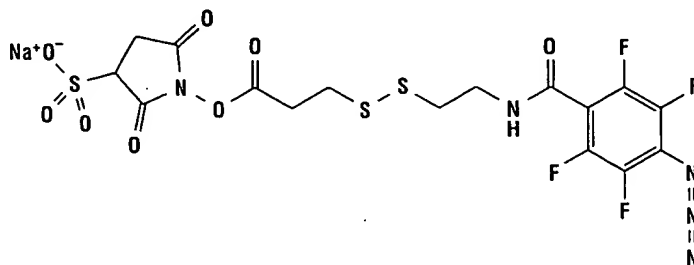
- Spacer maintains peptide-like character in the cross-linked species
- Resulting cross-link is susceptible to acid hydrolysis
- *Reactive groups:* NHS ester and bromoacetyl
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #73 (page 214)

Ordering Information

Product #	Description	Pkg Size	U.S. Price
22339ZZ	SBAP (Succinimidyl 3-(bromoacetamido)propionate)	50 mg	\$ 72

SFAD

Significantly improved photoconjugation efficiency over typical aryl azide-containing cross-linkers.



SFAD
M.W. 597.48
Spacer Arm 14.6 Å

Features/Benefits:

- Water-soluble; cleavable
- Perfluorophenyl azide moiety photolyzes at 320 nm
- Insertion efficiency approximately 70%
- Improved stability of the singlet perfluoroaryl nitrene reactive intermediate allows high-efficiency insertion with -CH bonds vs. low efficiency ring expansion with amine nucleophiles, typical of nonfluorinated aryl nitrenes
- ¹⁹F NMR can be used to monitor perfluoroaryl moiety transfer from one protein to another

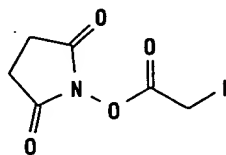
- *Reactive groups:* Sulfo-NHS ester and perfluoroaryl azide moiety
- *Reactive toward:* amino groups and -CH bonds
- Literature reference #'s 82, 83 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27719ZZ	SFAD (Sulfosuccinimidyl-[perfluoroazidobenzamido]-ethyl-1,3'-dithiopropionate)	50 mg	\$109

SIA

Suited to capturing amine and sulfhydryl groups in very close proximity.



SIA
M.W. 283.02
Spacer Arm 1.5 Å

Features/Benefits:

- Shortest sulfhydryl-reactive and amine-reactive heterobifunctional cross-linker available
- Non-cleavable; close proximity cross-linking agent

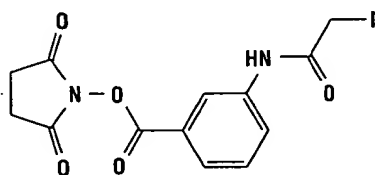
- NHS ester reacts with primary amines at pH 7-9 to form a stable amide bond
- Iodoacetyl group reacts with sulfhydryl groups at pH > 7.5 to form stable thioether bond
- *Reactive groups:* NHS ester and iodoacetyl
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 74, 75 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22349ZZ	SIA (N-Succinimidyl iodoacetate)	50 mg	\$ 39

SIAB

Popular alternative for making enzyme-antibody conjugates; reactive toward amines and sulfhydryls.



SIAB
M.W. 402.14
Spacer Arm 10.6 Å

Features/Benefits:

- Used to prepare stable enzyme-IgG conjugates
- Forms conjugates with liposomes

- *Reactive groups:* iodoacetyl and NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 33, 34 (page 213)

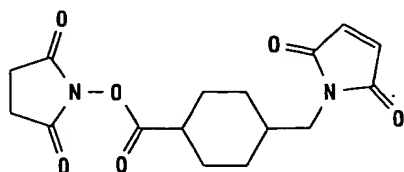
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22329ZZ	SIAB* (N-Succinimidyl[4-iodoacetyl]aminobenzoate)	50 mg	\$ 89

*Sulfonated, water-soluble analog also available; see Sulfo-SIAB.
See also: SIA, SBAP.

SMCC

Provides stable activated proteins.



SMCC
M.W. 334.32
Spacer Arm 11.6 Å

Features/Benefits:

- Cyclohexane bridge gives extra stability to the maleimide-reactive group; N-(4-carboxycyclohexylmethyl) maleimide groups are stable for 64 hours (in 0.1 M sodium phosphate buffer, pH 7.0 at 4°C)

- Ideal for coupling enzymes to antibodies; both the enzyme activity and antibody specificity can be preserved after coupling
- *Reactive groups:* NHS ester and maleimide
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 35, 52, 84, 89 (pages 213-214)

Ordering Information

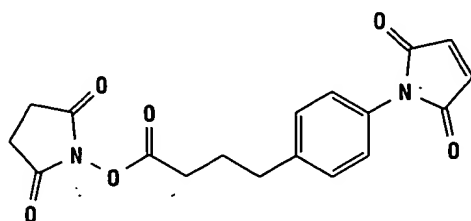
Product #	Description	Pkg. Size	U.S. Price
22360ZZ	SMCC* (Succinimidyl 4-[(N-maleimidomethyl)-cyclohexane-1-carboxylate])	50 mg	\$ 54

*Sulfonated, water-soluble analog also available; see Sulfo-SMCC.
See also: AMAS, BMPS, MBS, SMPB, SMPH.

800-874-3723

SMPB

Features extended chain length to limit steric hindrance.



SMPB
M.W. 356.33
Spacer Arm 11.6 Å

Features/Benefits:

- Extended chain analog of MBS
- Conjugates formed with SMPB were shown to be more stable in serum than SPDP conjugates

- *Reactive groups:* NHS ester and maleimide
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 36, 84 (pages 213-214)

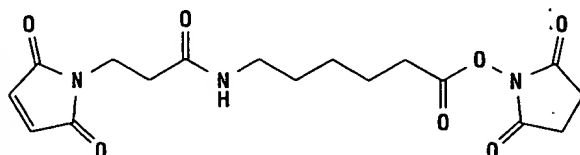
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22416ZZ	SMPB* (Succinimidyl 4-[p-maleimidophenyl]-butyrate)	50 mg	\$ 1

*Sulfonated, water-soluble analog also available; see Sulfo-SMPB.

SMPH

Spacer arm longer than that of SMPB, minimizing steric hindrance.



SMPH
M.W. 379.36
Spacer Arm 14.3 Å

Features/Benefits:

- Amine- and sulfhydryl-reactive heterobifunctional cross-linker
- NHS ester reacts with primary amines at pH 7-9 to form a stable amide bond

- Maleimide reacts with -SH groups at a pH of 6.5-7.5, forming stable thioether bond
- Non-cleavable; water-insoluble
- More hydrophilic character to the cross-bridge than BMPS, GMBS, EMCS or SMPB
- *Reactive groups:* maleimide and NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 74, 75 (page 214)

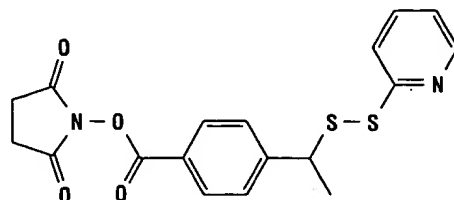
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22363ZZ	SMPH* (Succinimidyl-6-[(β-maleimidopropionamido)hexanoate])	50 mg	\$

*See also: BMPS, GMBS, EMCS, SMPB.

SMPT

Forms cleavable immunotoxins with greater stability in vivo.



SMPT
M.W. 388.46
Spacer Arm 20.0 Å

Features/Benefits:

- Contains a hindered disulfide bond; has formed immunotoxins with improved stability

- *In vitro*, an SMPT conjugate was as effective as conjugate formed with SPDP and 2-Iminoethanol
- Does not require exposing the antibody to reducing agents
- *Reactive groups:* NHS ester and pyridyldithio
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #37 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21558ZZ	SMPT* (4-Succinimidyltoxycarbonyl-methyl-α-[2-pyridyldithio]toluene)	50 mg	\$

*Sulfonated, long chain water-soluble analog also available; see Sulfo-LC-SMPT.

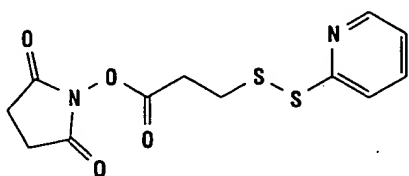
SPB

Amine-reactive DNA intercalating agent that has applications in cross-linking protein to DNA and photoimmobilizing compounds onto microwell plates.

See Product # 23013ZZ, (Succinimidyl-[4-(psoralen-8-yloxy)]butyrate), in Section 3-4, page 224.

SPDP

Classic heterobifunctional, cleavable cross-linker.



SPDP
M.W. 312.37
Spacer Arm 6.8 Å

Features/Benefits:

- Widely used in immunochemistry; conjugates used in drug carrier systems, antibody production and enzyme immunoassays have been successfully prepared with SPDP

- SPDP can be used as a protein thiolation reagent, resulting in available -SH groups
- *Reactive groups:* NHS ester and pyridyldithio
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 38, 86 (pages 213-214)

Ordering Information

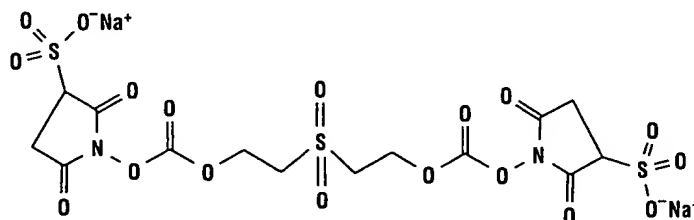
Product #	Description	Pkg. Size	U.S. Price
21857ZZ	SPDP* (N-Succinimidyl 3-[2-pyridyldithio]propionate)	50 mg	\$ 85

*See: LC-SPDP, Sulfo-LC-SPDP.

See also: SATA, SATP (protein thiolation reagents).

Sulfo-BSOCOES

Water-soluble, base-reversible analog of BSOCOES.



Sulfo-BSOCOES
M.W. 640.44
Spacer Arm 13.0 Å

Features/Benefits:

- Water-soluble
- Base-cleavable (pH 11.6, 2 hours, 37°C)
- *Reactive groups:* Sulfo-NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #39 (page 213)

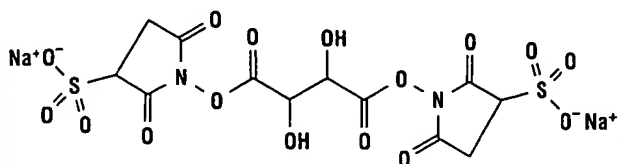
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21556ZZ	Sulfo-BSOCOES (Bis[2-(Sulfosuccinimidooxycarbonyloxy)-ethyl]sulfone)	50 mg	\$103

800-874-3723

Sulfo-DST

Water-soluble analog of DST; cleavable by oxidizing agents.



Sulfo-DST
M.W. 548.32
Spacer Arm 6.4 Å

- *Reactive groups:* Sulfo-NHS ester (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #40 (page 213)

Ordering Information:

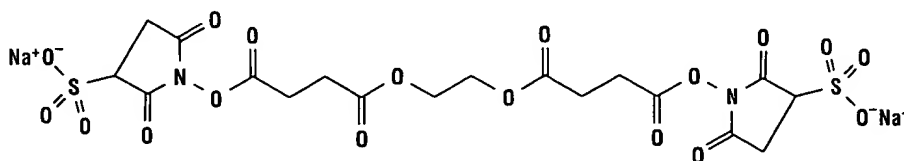
Product #	Description	Pkg. Size	U.S. Price
20591ZZ	Sulfo-DST (Disulfosuccinimidyl tartrate)	50 mg	\$ 98

Features/Benefits:

- Ideal for applications in which a cross-link reversibility is desired without disturbing protein S-S bonds

Sulfo-EGS

Water-soluble and cleavable – without harsh reducing agents.



Sulfo-EGS
M.W. 660.45
Spacer Arm 16.1 Å

Features/Benefits:

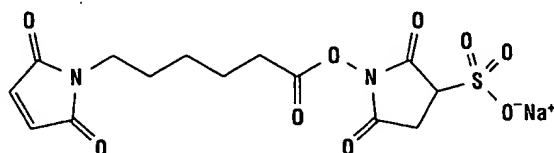
- Cross-links formed are cleavable at pH 8.5 using hydroxylamine for three to six hours at 37°C
- Lactose dehydrogenase retained 60% of its activity after reversible cross-linking with EGS
- *Reactive groups:* Sulfo-NHS esters (homobifunctional)
- *Reactive toward:* amino groups
- Literature reference #41 (page 213)

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
21566ZZ	Sulfo-EGS (Ethylene glycol bis[sulfosuccinimidylsuccinate])	50 mg	\$ 95

Sulfo-EMCS

Water-soluble, low immunogenicity and a little more reach.



Sulfo-EMCS
M.W. 410.33
Spacer Arm 9.4 Å

- Non-cleavable
- Increased sphere of coupling vs. Sulfo-GMBS
- Aliphatic spacer offers low potential for eliciting an immune response
- *Reactive groups:* maleimide and Sulfo-NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 23, 68, 69 (pages 213-214)

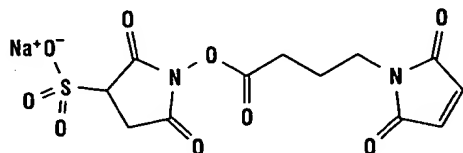
Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
22307ZZ	Sulfo-EMCS* (N-[ε-Maleimidocaproyloxy]sulfosuccinimide ester)	50 mg	\$14!

*Non-sulfonated, water-insoluble analog also available; see EMCS. See also: Sulfo-GMBS and Sulfo-KMUS.

Sulfo-GMBS

Sulfonated analog of GMBS offers water solubility, enhanced stability with less immunogenicity.



Sulfo-GMBS
M.W. 382.28
Spacer Arm 6.8 Å

Features/Benefits:

- Non-cleavable, membrane-impermeable
- Has a low potential for eliciting an immune response, ensuring that the primary response to the antigen-carrier protein conjugate is not diluted by a response against a determinant on the cross-linker

- GMBS and Sulfo-GMBS are reported to be less immunogenic than SMCC
- Sulfo-GMBS has improved solubility in water and aqueous buffer systems
- *Reactive groups:* maleimide and Sulfo-NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #23 (page 213)

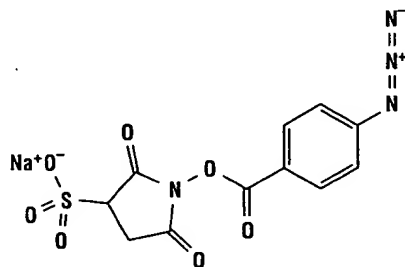
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22324ZZ	Sulfo-GMBS* (N-[γ-Maleimidobutyryloxy]sulfo-succinimide ester)	50 mg	\$137

*See also: Sulfo-EMCS, Sulfo-KMUS, Sulfo-MBS.

Sulfo-HSAB

Water-soluble, amine-reactive, photoreactive cross-linker.



Sulfo-HSAB
M.W. 362.25
Spacer Arm 9.0 Å

Features/Benefits:

- Useful for the preparation of conjugates for the purpose of producing antibodies

- *Reactive groups:* phenyl azide and Sulfo-NHS ester
- *Reactive toward:* amino groups
- Literature reference #'s 24, 30 (page 213)

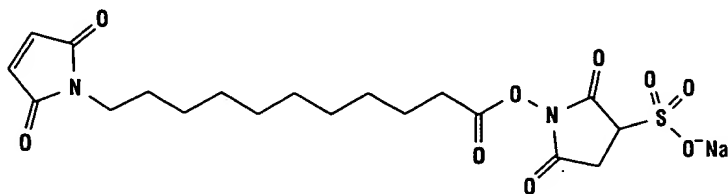
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21563ZZ	Sulfo-HSAB (N-Hydroxysulfosuccinimidyl-4-azidobenzoate)	50 mg	\$ 54

800-874-3723

Sulfo-KMUS

Water-soluble, long chain heterobifunctional cross-linker.



Sulfo-KMUS
M.W. 480.47
Spacer Arm 15.7 Å

Features/Benefits:

- Non-cleavable extended aliphatic cross-bridge
- Water-soluble with enhanced sphere of coupling
- Sulfo-NHS ester reacts with primary amines at pH 7-9 to form a stable amide bond
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming a stable thioether bond
- *Reactive groups:* maleimide and Sulfo-NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #23 (page 213)

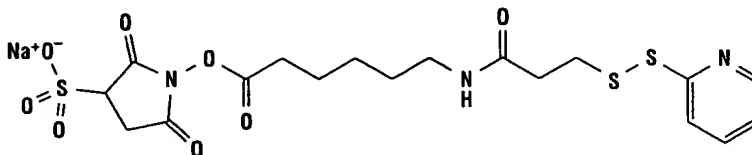
Ordering Information

Product #	Description	Pkg Size	U.S. Price
21111ZZ	Sulfo-KMUS* (N-{ κ -Maleimidoundecanoyloxy}-sulfosuccinimide ester)	50 mg	\$159

*See also: Sulfo-GMBS, Sulfo-EMCS.

Sulfo-LC-SPDP

Classic water-soluble, heterobifunctional, cleavable cross-linker.



Sulfo-LC-SPDP
M.W. 527.57
Spacer Arm 15.6 Å

Features/Benefits:

- Sulfo-LC-SPDP releases a detectable byproduct after reacting with free sulfhydryl groups; by measuring the release of pyridine-2-thione at 343 nm, the reaction can be easily followed
- *Reactive groups:* pyridylthio and Sulfo-NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 38, 85 (pages 213-214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21650ZZ	Sulfo-LC-SPDP (Sulfosuccinimidyl 6-[3'-(2-pyridylthio)-propionamido]hexanoate)	50 mg	\$257

3

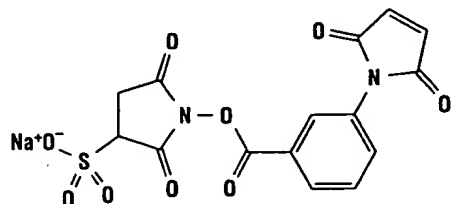
member <http://www.merck.com>

3-7

200

Sulfo-MBS

Useful for coupling proteins, enzymes to antibodies, toxins to antibodies, and haptens to carrier proteins.



Sulfo-MBS
M.W. 416.30
Spacer Arm 9.9 Å

- *Reactive groups:* Sulfo-NHS ester and maleimide
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #42 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22312ZZ	Sulfo-MBS* (<i>m</i> -Maleimidobenzoyl- <i>N</i> -hydroxysulfo-succinimide ester)	50 mg	\$ 94

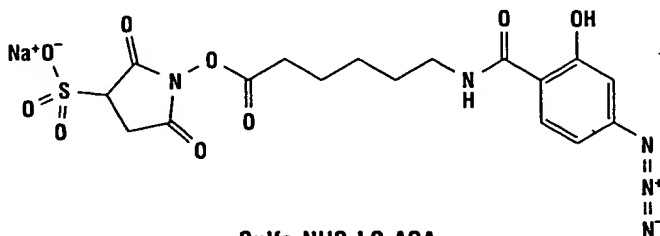
*See also: Sulfo-EMCS, Sulfo-GMBS, Sulfo-KMUS.

Features/Benefits:

- Water-soluble; non-cleavable
- Membrane-impermeable

Sulfo-NHS-LC-ASA

An ¹²⁵I label can be easily and effectively incorporated into this reagent before the acylation step and used to radiolabel conjugates.



Sulfo-NHS-LC-ASA
M.W. 491.41
Spacer Arm 18.0 Å

Features/Benefits:

- Photolysis reaction is readily initiated by long wave UV light
- Simplifies detection of photoaffinity labeled complexes and the determination of cross-linked loci
- Sulfo-NHS-LC-ASA is water-soluble, and has a long spacer arm to overcome steric restraints
- *Reactive groups:* hydroxy phenyl azide and Sulfo-NHS ester
- *Reactive toward:* amino groups
- Literature reference #27 (page 213)

Ordering Information

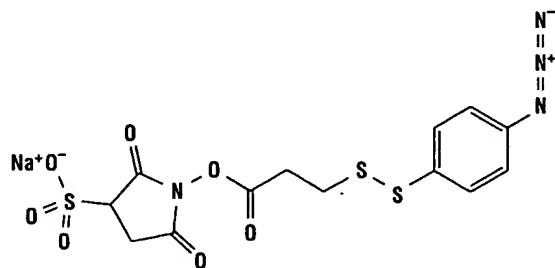
Product #	Description	Pkg. Size	U.S. Price
27735ZZ	Sulfo-NHS-LC-ASA* (Sulfosuccinimidyl[4-azidosalicylamido]-hexanoate)	50 mg	\$225

*See also: SASD.

800-874-3723

Sulfo-SADP

Highly versatile, water-soluble, cleavable and photoreactive cross-linker.



Sulfo-SADP
M.W. 454.44
Spacer Arm 13.9 Å

Features/Benefits:

- Cleavable by 50 mM dithiothreitol, 100 mM β-mercaptoethanol or 1% sodium borohydride

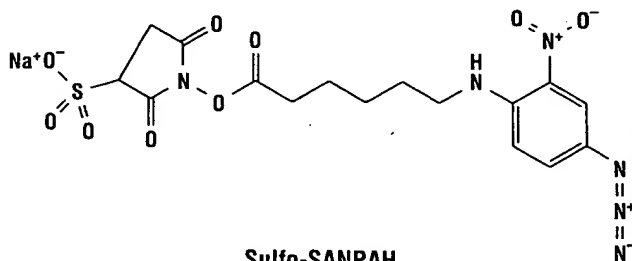
- Photolysis is achieved by irradiation at 265-275 nm
- *Reactive groups:* phenyl azide and Sulfo-NHS ester
- *Reactive toward:* amino groups
- Literature reference #20 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21553ZZ	Sulfo-SADP (Sulfosuccinimidyl[4-azidophenylthio]-propionate)	50 mg	\$121

Sulfo-SANPAH

Extended chain length, photoactivatable cross-linker.



Sulfo-SANPAH
M.W. 492.40
Spacer Arm 18.2 Å

Features/Benefits:

- Optimal photolysis occurs at 320-350 nm, a condition that limits damage to biomolecules by irradiation

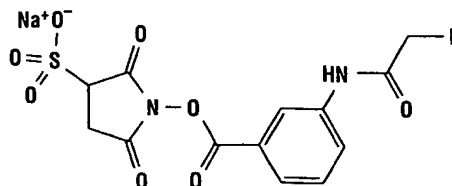
- Water-soluble; non-cleavable
- *Reactive groups:* nitrophenyl azide and Sulfo-NHS ester
- *Reactive toward:* amino groups
- Literature reference #30 (page 213)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22589ZZ	Sulfo-SANPAH (Sulfosuccinimidyl 6-[4'-azido-2'-nitro-phenylamino]hexanoate)	50 mg	\$13

Sulfo-SIAB

Popular alternative for making enzyme-antibody conjugates - reactive toward amines and sulfhydryls.



Sulfo-SIAB
M.W. 504.19
Spacer Arm 10.6 Å

Features/Benefits:

- *Reactive groups:* iodoacetate and Sulfo-NHS ester
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #45 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22327ZZ	Sulfo-SIAB (Sulfosuccinimidyl[4-iodoacetyl]aminobenzoate)	50 mg	\$ 9

Special Product

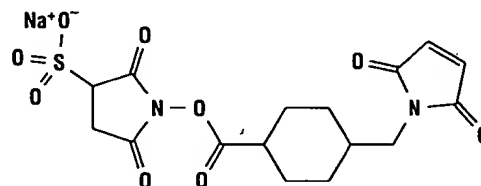


Sulfo-SMCC

Provides stable activated proteins.

Features/Benefits:

- Water-soluble, non-cleavable and membrane-impermeable
- Cyclohexane bridge gives extra stability to the maleimide-reactive group; *N*-(4-carboxycyclohexylmethyl)maleimide groups are stable for 64 hours (in 0.1 M sodium phosphate buffer, pH 7.0 at 4°C)
- Ideal for coupling enzymes to antibodies; both the enzyme activity and antibody specificity can be preserved after coupling
- *Reactive groups*: Sulfo-NHS ester and maleimide
- *Reactive toward*: amino and sulfhydryl groups
- Literature reference #46 (page 214)



Sulfo-SMCC
M.W. 436.37
Spacer Arm 11.6 Å

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22322ZZ	Sulfo-SMCC (Sulfosuccinimidyl 4-[<i>N</i> -maleimidomethyl]-cyclohexane-1-carboxylate)	50 mg	\$104

3

PIERCE

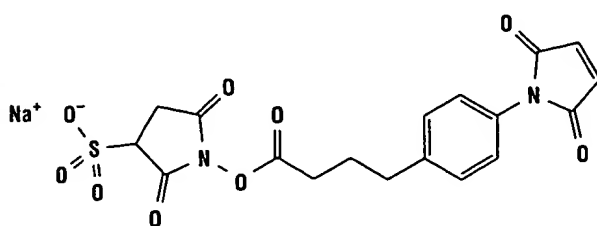
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800-874-3723

Sulfo-SMPB

Extended chain length analog of Sulfo-MBS.



Sulfo-SMPB
M.W. 458.38
Spacer Arm 14.5 Å

Features/Benefits:

- Extended chain length limits steric hindrance
- Water-soluble; non-cleavable

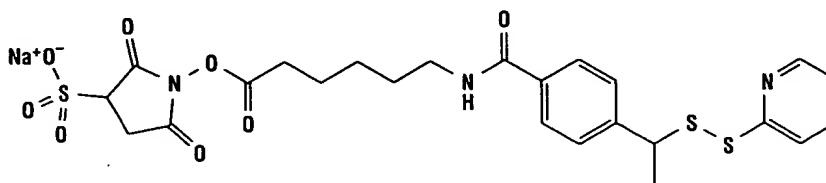
- Membrane-impermeable
- *Reactive groups:* Sulfo-NHS ester and maleimide
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #'s 36, 47 (pages 213-214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22317ZZ	Sulfo-SMPB (Sulfosuccinimidyl 4-[p-maleimidophenyl]-butyrate)	50 mg	\$101

Sulfo-LC-SMPT

Form cleavable immunotoxins with greater stability in vivo.



Sulfo-LC-SMPT
M.W. 603.67
Spacer Arm 20.0 Å

Features/Benefits:

- Contains a hindered disulfide bond; has formed immunotoxins with improved stability
- *In vitro*, SMPT conjugates are as effective as conjugates formed with SPDP and 2-Iminoethanol
- Does not require exposing the antibody to reducing agents
- Offers an extended spacer arm and water solubility

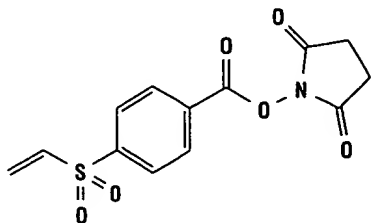
- *Reactive groups:* Sulfo-NHS ester and pyridyldithio
- *Reactive toward:* amino and sulfhydryl groups
- Literature reference #48 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
21568ZZ	Sulfo-LC-SMPT (Sulfosuccinimidyl-6-[α-methyl-α-(2-pyridyldithio)toluamido]hexanoate)	50 mg	\$229

SVSB

Combines the advantages of the vinylsulfone-reactive group with those of the classical NHS ester.



SVSB
M.W. 309.30
Spacer Arm 8.3 Å

Features/Benefits:

- Novel heterobifunctional reagent containing an amine-reactive NHS and a sulfhydryl-reactive vinylsulfone
- Non-cleavable; water-insoluble
- NHS ester reacts with primary amines at pH 7-9 to form a stable amide bond

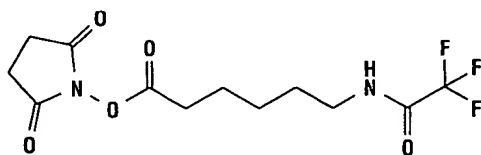
- Vinylsulfone reacts with -SH groups, forming a stable thioether linkage
- Conjugates prepared with vinylsulfone-reactive group form a single stereoisomer
- Unlike the maleimide active group, vinylsulfone is not subject to hydrolytic degradation in aqueous environments
- After modification of an amine-containing molecule with SVSB, the vinylsulfone intermediate can be stored for long periods without loss of sulfhydryl reactivity
- *Reactive groups:* vinylsulfone and NHS ester
- *Reactive toward:* sulfhydryl and amino groups
- Literature reference #'s 78-80 (page 214)

Ordering Information

Product #	Description	Pkg Size	U.S. Price
22358ZZ	SVSB (N-Succinimidyl-4-vinylsulfonyl)benzoate	50 mg	\$107

TFCS

Amine-reactive cross-linker with a latent amino group available on demand.



TFCS
M.W. 324.25
Spacer Arm 7.7 Å

Features/Benefits:

- Amine-reactive modification agent with a protected primary amine group
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds
- Used to extend lysine side-chain length to reduce steric hindrance

- Temporarily block amine groups in target molecules
- Trifluoroacetyl protecting group released by phosphate or borate buffer, pH 7.8-8.1
- Unmasked amino group ready for reaction with any amine-reactive cross-linker for conjugate preparation
- Non-cleavable; water-insoluble
- *Reactive groups:* NHS ester and trifluoroacetyl-protected -NH₂
- *Reactive toward:* amino groups and NHS esters/Sulfo-NHS esters or EDC-activated carboxyl groups

Ordering Information

Product #	Description	Pkg Size	U.S. Price
22299ZZ	TFCS (N-[(2-Trifluoroacetyl)caproyloxy]succinimide ester)	50 mg	\$ 39

Special Product

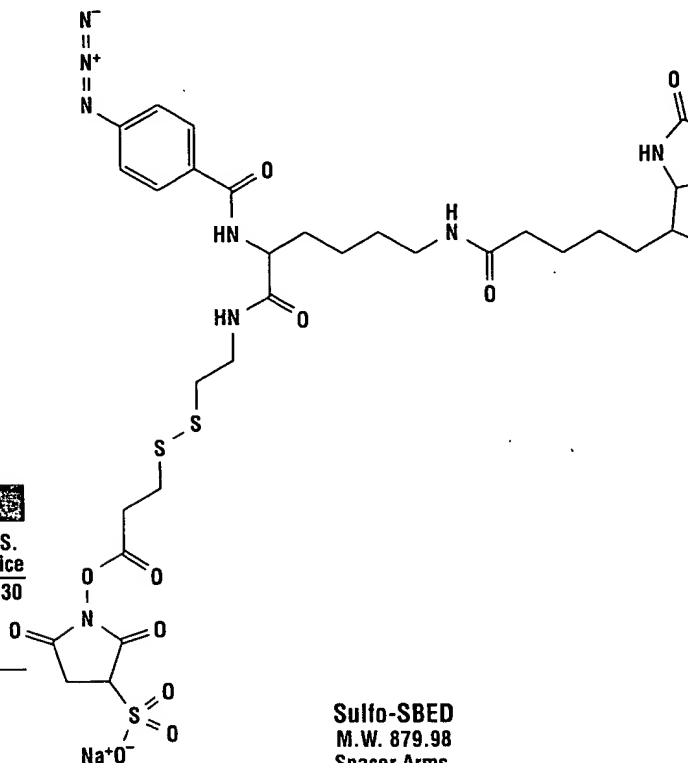


Features/Benefits:

- Amine group-specific reactivity
- Nonspecific photoreactivity
- A biotin handle; thiol-cleavable
- Soluble in DMF, DMSO and MeOH
- Moisture and light sensitive
- *Reactive groups:* phenyl azide, Sulfo-NHS ester and biotin
- *Reactive toward:* amino groups and avidin/streptavidin/NeutrAvidin™ Biotin-Binding Protein
- Literature reference #'s 49, 50, 87, 91 (page 214)

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
33033ZZ	Sulfo-SBED (Sulfosuccinimidyl [2-6-(biotinamido)-2-(p-azidobenzamido)-hexanoamido]ethyl-1,3'-dithiopropionate)	10 mg	\$130

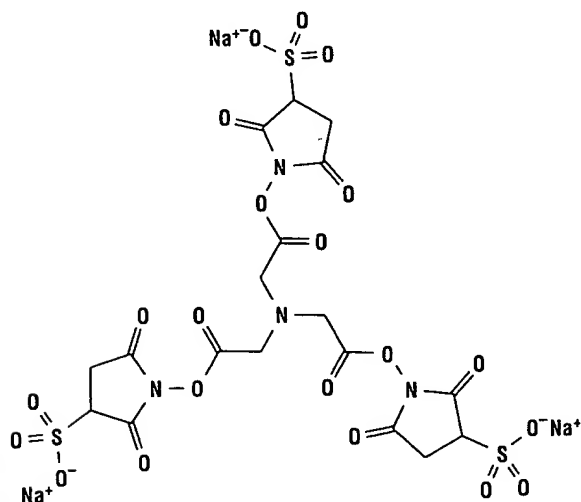


Sulfo-SBED
M.W. 879.98
Spacer Arms

Sulfo-NHS ester 13.7 Å
Phenyl azide 9.1 Å
Biotin 19.1 Å

Sulfo-TSAT

Water-soluble, amine-reactive agent for preparing trimeric aggregates.



Sulfo-TSAT
M.W. 788.49
Spacer Arm 4.2 Å

Features/Benefits:

- Novel amine-reactive trifunctional cross-linking agent
- Sulfo-NHS ester couples to amines at pH 7-9 to form stable amide bonds
- Useful for the preparation of multicomponent aggregates
- Non-cleavable; water-soluble
- Literature in nearest neighbor studies
- Core molecule for the construction of dendritic polymers
- *Reactive group:* Sulfo-NHS ester
- *Reactive toward:* amino groups

Ordering Information

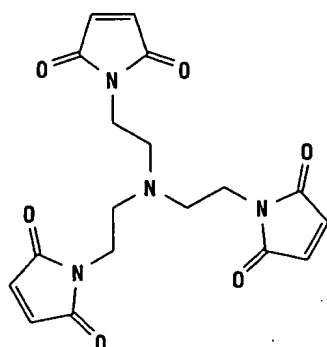
Product #	Description	Pkg. Size	U.S. Price
33053ZZ	Sulfo-TSAT (Tris-sulfosuccinimidyl aminotriacetate)	50 mg	\$12

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TMEA

Sulfhydryl-reactive tool for preparing trimeric aggregates.



TMEA
M.W. 386.36
Spacer Arm 10.3 Å

Features/Benefits:

- Novel sulfhydryl-reactive trifunctional cross-linking agent
- Maleimides react with -SH groups at a pH of 6.5-7.5, forming stable thioether linkages
- Useful for the preparation of trimeric complexes of cysteine-containing peptides and other thiol-containing compounds
- Non-cleavable; water-insoluble
- Application in nearest neighbor studies
- Core molecule for the construction of dendritic polymers
- *Reactive group:* maleimide
- *Reactive toward:* sulfhydryl groups

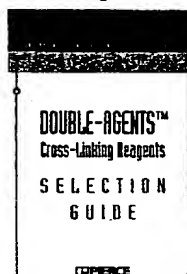
Ordering Information

Product #	Description	Pkg Size	U.S. Price
33043ZZ	TMEA (Tris-[2-maleimidoethyl]amine)	50 mg	\$109

Cross-linking Reagent Buffer Scheme

Type of Cross-linker	Buffer
Imidoester	BupH™ Borate Buffer Packs, pH 8.5 (Product # 28384ZZ) or BupH™ Carbonate-Bicarbonate Buffer Packs, pH 9.0 (Product # 28382ZZ)
Homobifunctional NHS-Ester Cross-linking Buffer	BupH™ Phosphate Buffered Saline Packs, pH 7.2 (Product # 28372ZZ) or BupH™ Modified Dulbecco's Phosphate Buffered Saline Packs, pH 7.4 (Product # 28374ZZ)
Heterobifunctional NHS-Ester Maleimide	BupH™ Phosphate Buffered Saline Packs, pH 7.2 (Product # 28372ZZ) or BupH™ Modified Dulbecco's Phosphate Buffered Saline Packs, pH 7.4 (Product # 28374ZZ)
Carbodiimide	BupH™ MES Buffered Saline, pH 4.7 (Product # 28390ZZ)
EDC with NHS or Sulfo-NHS	BupH™ Phosphate Buffered Saline Packs, pH 7.2 (Product # 28372ZZ) or BupH™ Modified Dulbecco's Phosphate Buffered Saline Packs, pH 7.4 (Product # 28374ZZ)

Double-Agents™ Cross-Linking Reagents Selection Guide



Contact Pierce, the world leader in cross-linking reagents for life science applications, for a free copy of the **Double-Agents™ Cross-Linking Reagents Selection Guide**. Written for both the novice and the expert, this guide helps you select the best reagent(s) based on your criteria and intended application. Having helped you narrow your choices, the guide provides other relevant information (molecular weight, spacer arm length, structure, etc.) to further help you select a reagent.

Ordering Information

Product #	Description
1600250	Double-Agents™ Cross-Linking Reagents Selection Guide

In a hurry to select a cross-linker? Log on to the Pierce web site at www.piercenet.com and use our online selection guide.

800-874-3723

Pierce Cross-linkers at a Glance

Double-Agents™ Cross-linker Product # Acronym	Reactive Toward						Cleavable By				Iodina- table		Latent Functiona Group
	-NH ₂ Amines	-SH Sulf- hydryls	Carbo- hydrates	Nonselec- tive (photo- reactive)	-COOH Carboxyls	-OH Hydroxyl	Thiols	Base	Periodate	Hydroxyl- amine	Yes	No	
21509ZZ ABH			X	X								X	
22101ZZ AEDP	X				X		X					X	
22295ZZ AMAS	X	X										X	
21451ZZ ANB-NOS	X			X								X	
27720ZZ APDP		X		X			X				X		
20108ZZ APG				X								X	
21512ZZ ASBA				X	X						X		
21564ZZ BASED				X			X				X		
22331ZZ BMB		X										X	
22332ZZ BMDB		X							X			X	
22330ZZ BMH		X										X	
22323ZZ BMOE		X										X	
22296ZZ BMPA	X	X										X	
22297ZZ BMPH		X	X									X	
22298ZZ BMPS	X	X										X	
22336ZZ BM(PEO) ₃		X										X	
22337ZZ BM(PEO) ₄		X										X	
21600ZZ BSOCES	X							X				X	
21580ZZ BS'	X											X	
21525ZZ DFDNB	X											X	
20663ZZ DMA	X											X	
21666ZZ DMP	X											X	
20700ZZ DMS	X											X	
21702ZZ DPDPB		X					X					X	
20593ZZ DSG	X											X	
22585ZZ DSP	X						X					X	
21555ZZ DSS	X											X	
20589ZZ DST	X								X			X	
20665ZZ DT8P	X						X					X	
22335ZZ DTME		X					X					X	
21578ZZ DTSSP	X						X					X	
22980ZZ EDC	X				X							X	
21565ZZ EGS	X									X		X	
22306ZZ EMCA	X	X										X	
22106ZZ EMCH		X	X									X	
22308ZZ EMCS	X	X										X	
22309ZZ GMBS	X	X										X	
22334ZZ HBVS		X										X	
22211ZZ KMUA	X	X										X	
22111ZZ KMUH		X	X									X	
22362ZZ LC-SMCC	X	X										X	
21651ZZ LC-SPDP	X	X					X					X	
22311ZZ MBS	X	X										X	

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http://www.pierce.com

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Cross-linking Reagents

Double-Agents™ Cross-linker Product # Acronym	Reactive Toward						Cleavable By				Iodina- table		Latent Functional Group
	-NH ₂ Amines	-SH Sulf- hydriyls	Carbo- hydrates	Nonselec- tive (photo- reactive)	-COOH Carboxyls	-OH Hydroxyl	Thiols	Base	Periodate	Hydroxyl- amine	Yes	No	
22303ZZ M ₂ C ₂ H		X	X									X	
22305ZZ MPBH		X	X									X	
22605ZZ MSA	X											X	-COOH
27714ZZ NHS-ASA	X			X							X		
22301ZZ PDPH		X	X				X					X	
28100ZZ PMPI		X				X						X	
21533ZZ SADP	X			X			X					X	
33030ZZ SAED	X			X			X					X	
21549ZZ SAND	X			X			X					X	
22600ZZ SANPAH	X			X								X	
27716ZZ SASD	X			X			X				X		
26102ZZ SATA	X	X										X	-SH
26100ZZ SATP	X	X										X	-SH
22339ZZ SBAP	X	X										X	
27719ZZ SFAD	X			X			X					X	
22349ZZ SIA	X	X										X	
22329ZZ SIAB	X	X										X	
22360ZZ SMCC	X	X										X	
22416ZZ SMPB	X	X										X	
22363ZZ SMPH	X	X										X	
21558ZZ SMPT	X	X					X					X	
21857ZZ SPDP	X	X					X					X	
21556ZZ Sulfo-BSOCOES	X							X				X	
20591ZZ Sulfo-DST	X								X			X	
21566ZZ Sulfo-EGS	X									X		X	
22307ZZ Sulfo-EMCS	X	X										X	
22324ZZ Sulfo-GMBS	X	X										X	
21563ZZ Sulfo-HSAB	X			X								X	
21111ZZ Sulfo-KMUS	X	X										X	
21650ZZ Sulfo-LC-SPDP	X	X					X					X	
22312ZZ Sulfo-MBS	X	X										X	
27735ZZ Sulfo-NHS-LC-ASA	X			X							X		
21553ZZ Sulfo-SADP	X			X			X					X	
22589ZZ Sulfo-SANPAH	X			X								X	
22327ZZ Sulfo-SIAB	X	X										X	
22322ZZ Sulfo-SMCC	X	X										X	
22317ZZ Sulfo-SMPB	X	X										X	
21568ZZ Sulfo-LC-SMPT	X	X					X					X	
33033ZZ Sulfo-SBED*	X			X			X					X	
33053ZZ Sulfo-TSAT**	X											X	
22358ZZ SVSB	X	X										X	
22299ZZ TFCS	X				X								-NH ₂
33043ZZ TMEA**		X										X	

*Trifunctional cross-linking reagent; binds to Avidin, Streptavidin and NeutrAvidin™ Biotin-Binding Protein.

**Trifunctional cross-linking agent.

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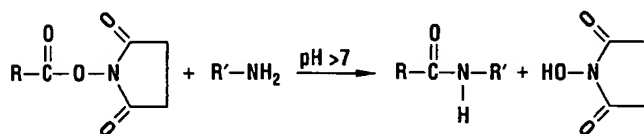
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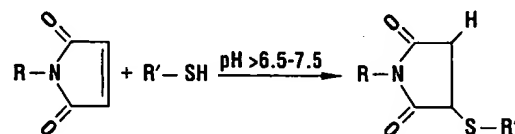
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Active Group Reaction Schemes

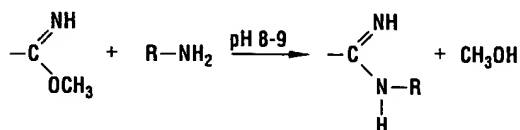
NHS-Ester Reaction Scheme



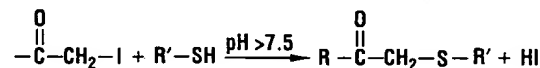
Maleimide Reaction Scheme



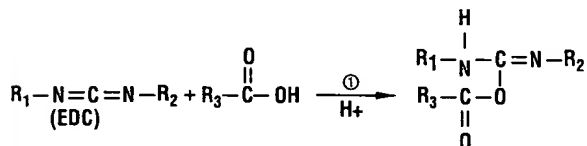
Imidoester Reaction Scheme



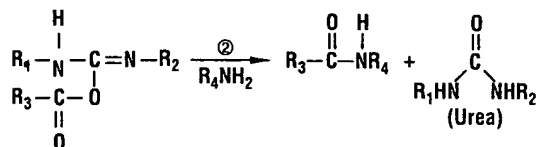
Active Halogen Reaction Scheme



EDC Coupling Reaction Scheme

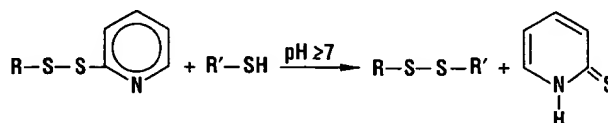


EDC reacts with carboxylic acid group and activates the carboxyl group, allowing it to be coupled to the amino group (R_4NH_2) in the reaction mixture.



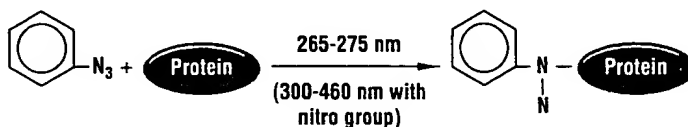
EDC is released as a soluble urea derivative after displacement by the nucleophile, R_4NH_2 .

Pyridyl Disulfide Reaction Scheme

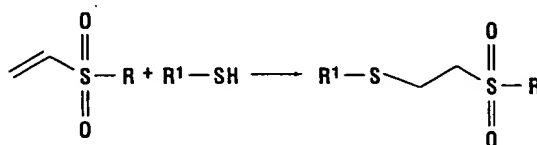


Active Group Reaction Schemes

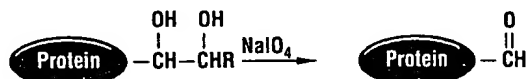
Azidophenyl Photolysis



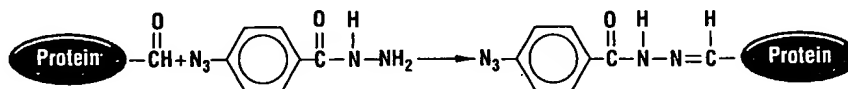
Vinyl-Sulfone Reaction Scheme



Hydrazide Reaction Scheme



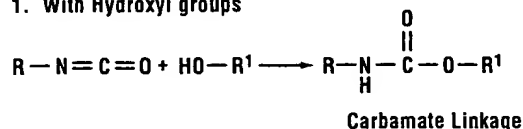
The oxidation of a Protein Carbohydrate (*cis*-diol) to an aldehyde.



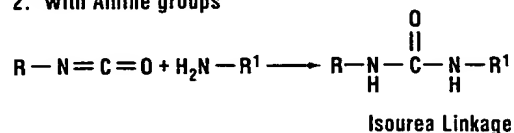
ABH, or Azidobenzoyl Hydrazide, reacts with the aldehyde on the protein to form an arylazide activated protein.

Isocyanate Reaction Schemes

1. With Hydroxyl groups



2. With Amine groups

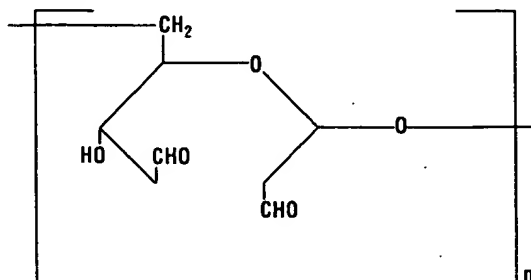


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Activated Dextran Coupling Kits

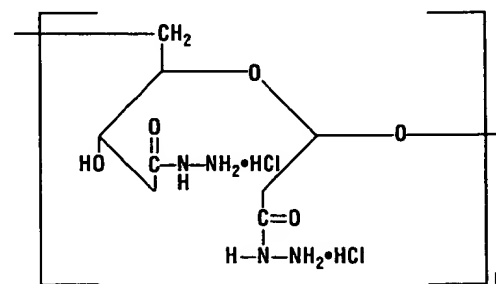
Novel water-soluble reagents capable of multimeric aggregate formation.

Couple amine-containing ligands and biomolecules



Aldehyde-Activated Dextran
Average M.W. (dextran) 40 kD

Couple aldehyde- or carboxyl-containing ligands and biomolecules



Hydrazide-Activated Dextran
Average M.W. (dextran) 40 kD

Potential Applications:

- Coupling both high and low molecular weight biomolecules to a single soluble matrix
- Serving as a water-soluble scavenger of proteases from cell preparations
- Isolating a target antigen from solution containing dextran-coupled antibodies
- Isolating a target antibody from solution via a specific ligand
- Stabilizing enzymes for research or process-level applications
- Forming antibody-enzyme conjugates for use in immunoassay development
- Preparing immunoconjugates
- Acting as a poly-functional cross-linking agent to capture multicomponent systems
- Improving the solubility properties of isolated membrane proteins

Reference

1. Mallia, A.K. and Vigna, R.A. (1998). Nonimmunogenic-activated carriers: aldehyde-activated and hydrazide-activated dextran. *Previews* 1(4), 16-17.

Ordering Information

Product #	Description	Pkg Size	U.S. Price
20890ZZ	Aldehyde-Activated Dextran Coupling Kit	Kit	\$ 97
	Includes: Aldehyde-Activated Dextran* [CHO Loading: ~300 moles/mole of dextran]	5 x 5 mg	
	Sodium Cyanoborohydride	190 mg	
	BupH™ Phosphate Buffered Saline	1 pack	
20900ZZ	Hydrazide-Activated Dextran Coupling Kit	Kit	\$127
	Includes: Hydrazide-Activated Dextran* [Hydrazide Loading: 1-2 μmoles hydrazide/mg dextran]	5 x 5 mg	
	Sodium Cyanoborohydride	190 mg	
	BupH™ Phosphate Buffered Saline	1 pack	

*The average molecular weight of dextran used in these preparations is 40 kD.

Compatible Products

22980ZZ	EDC (1-Ethyl-3-[3-Dimethylaminopropyl] carbodiimide Hydrochloride)	5 gm	\$ 61
22981ZZ	EDC	25 gm	\$198
28372ZZ	BupH™ Phosphate Buffered Saline Packs Each pack yields 500 ml of 0.1 M phosphate, 0.15 M NaCl, pH 7.2 when dissolved in 500 ml of distilled water.	40 packs	\$ 83
44892ZZ	AminoLink® Reductant (Sodium Cyanoborohydride)	2 gm	\$ 25

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Internet: <http://www.pierce.com>

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Pierce Cross-linking Reagent Literature References

- O'Shannessy, D.J. and Quarles, R.H. (1985). Specific conjugation reactions of the oligosaccharide moieties of immunoglobulins. *J. Applied Biochem.* 7, 347-355.
- Krieg, U.C., Walter, P. and Johnson, A.E. (1986). Photocross-linking of the signal sequence of nascent preprolactin to the 54-kilodalton polypeptide of the signal recognition particle. *Proc. Natl. Acad. Sci. USA* 83, 8604-8606.
- Traut, R.R., et al. (1989). *Protein Function, A Practical Approach*. Oxford: IRL Press, p. 101.
- Sgro, J., Jacrot, B. and Chroboczek, J. (1986). *Eur. J. Biochem.* 154, 69-76.
- Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 284, 416.
- Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 214, 416.
- Chen, L.L., Rosa, J.J., Turner, S. and Pepinsky, R.B. (1991). Production of multimeric forms of CD4 through a sugar-based cross-linking strategy. *J. Biol. Chem.* 266(27), 18237-18243.
- Bouzar, Z., Fouchereau-Person, M., Taboulet, J., Moulhtar, M.S. and Milhaud, G. (1986). Purification and characterization of calcitonin receptors in rat kidney membranes by covalent cross-linking techniques. *Eur. J. Biochem.* 155, 141-147.
- Knoller, S., Shpungin, S. and Pick, E. (1991). The membrane-associated component of the amphiphile-activated, cytosol-dependent superoxide-forming NADPH oxidase of macrophages is identical to cytochrome b559. *J. Biol. Chem.* 266, 2795-2804.
- Kornblatt, J.A. and Lake, D.F. (1980). Cross-linking of cytochrome oxidase subunits with difluorodinitrobenzene. *Can. J. Biochem.* 58, 219-224.
- Hartman, F.C. and Wold, F. (1967). Cross-linking of bovine pancreatic ribonuclease A with dimethyl adipimidate. *Biochem.* 6(8), 2439-2448.
- Schneider, C., Newman, R.A., Sutherland, D.R., Asser, U. and Greaves, M.F. (1982). A one-step purification of membrane proteins using a high efficiency immunomatrix. *J. Biol. Chem.* 257(18), 10766-10769.
- Wang, D. and Moore, S. (1977). Polyspermine-ribonuclease prepared by cross-linkage with dimethyl suberimidate. *Biochem.* 16(13), 2937-2941.
- Chen, L.L., Frankel, A.D., Harder, J.L., Fawell, S., Barsoun, J. and Pepinsky, B. (1995). Increased cellular uptake of the human immunodeficiency virus-1 tat protein after modification with biotin. *Anal. Biochem.* 227, 168-175.
- Waugh, S.M., DiBella, E.E. and Pilch, P.F. (1989). Isolation of a proteolytically derived domain of the insulin receptor containing the major site of cross-linking/binding. *Biochemistry* 28, 3448-3455. (EGS example)
- Joshi, S. and Burrows, R. (1990). AT synthase complex from bovine heart mitochondria. *J. Biol. Chem.* 265, 14518-14525.
- Cox, G.W., Mattieson, B.J., Giardina, S.L. and Varesio, L. (1990). Characterization of IL-2 receptor expression and function on murine macrophages. *J. Immunol.* 145, 1719-1726.
- Farries, T.C. and Atkinson, J.P. (1989). Biosynthesis of properdin. *J. Immunol.* 142, 842-847.
- Shivdasani, R.A. and Thomas, D.W. (1988). Molecular associations of IA antigens after T-B cell interactions. *J. Immunol.* 141, 1252-1260.
- Jung, S.M. and Moroi, M. (1983). Cross-linking of platelet glycoprotein Ib by N-succinimidyl(4-azidophenylthio)propionate and 3,3'-dithiobis(sulfosuccinimidyl propionate). *Biochem. Biophys. Acta* 761, 152-162.
- Taniuchi, M., Clark, H.B. and Johnson, Jr., E.M. (1986). Induction of nerve growth factor receptor in Schwann cells after axotomy. *Proc. Natl. Acad. Sci. USA* 83, 4094-4098.
- Millar, J.B. and Rozengur, E. (1990). Chronic desensitization to bombesin by progressive down-regulation of bombesin receptors in Swiss 3T3 cells. *J. Biol. Chem.* 265, 12052-12058.
- Fujiwara, K., et al. (1988). Sandwich enzyme immunoassay of tumor-associated antigen sialosylated Lewis (x) using β -D-galactosidase coupled to a monoclonal antibody of IgM isotype. *J. Immunol. Meth.* 112, 77-83.
- Ballmer-Hofer, K., Schlup, V., Burn, P. and Burger, M.M. (1982). Isolation of *in situ* cross-linked ligand-receptor complexes using an anticross-linker specific antibody. *Anal. Biochem.* 126, 246-250.
- Kitagawa, T. and Aikawa, T. (1976). Enzyme coupled immunoassay of insulin using a novel coupling reagent. *J. Biochem. (Tokyo)* 79, 233-236.
- Chamow, S.M., Kogan, T.P., Peers, D.H., Hastings, R.C., Byrn, R.A. and Askenaszi, A. (1992). Conjugation of soluble CD4 without loss of biological activity via a novel carbohydrate-directed cross-linking reagent. *J. Biol. Chem.* 267(22), 15916-15922.
- van der Horst, G.T.J., Mancini, G.M.S., Brossmer, R., Rose, U. and Verheijen, F.W. (1990). Photoaffinity labeling of a bacterial sialidase with an aryl azide derivative of sialic acid. *J. Biol. Chem.* 265(19), 10801-10804. (NHS-ASA example)
- Greenfield, R.S., Kaneko, T., Daves, A., Edson, M.A., Fitzgerald, A., Olech, L.J., Grattan, J.A., Spitalny, G.L. and Braslawsky, G.R. (1990). Evaluation *in vitro* of adriamycin immunoconjugates synthesized using an acid sensitive hydrazone linker. *Cancer Res.* 50, 6600-6607.
- Lewis, R.V., Roberts, M.F., Dennis, E.A. and Allison, W.S. (1977). Photoactivated heterobifunctional cross-linking reagents which demonstrate the aggregation state of phospholipase A2. *Biochemistry* 16, 5650-5654. (ANB-NOS example)
- Wood, C.L. and O'Dorisio, M.S. (1985). Covalent cross-linking of vasoactive intestinal polypeptide to its receptors on intact human lymphoblasts. *J. Biol. Chem.* 260, 1243-1247. (HSAB example)
- Chattopadhyay, A., James, H.I. and Fair, D.S. (1992). Molecular recognition sites on factor Xa which participate in the prothrombinase complex. *J. Biol. Chem.* 267, 12323-12329.
- Kitagawa, T., Shimozono, T., Aikawa, T., Yoshida, T. and Nishimura, H. (1981). Preparation and characterization of heterobifunctional cross-linking reagents for protein modifications. *Chem. Pharm. Bull.* 29(4), 1130-1135.
- Cumber, A.J., Forrester, J.A., Foxwell, B.M.J., Ross, W.C.J. and Thorpe, P.W. (1985). Preparation of antibody-toxin conjugates. *Meth. Enzymol.*, New York: Academic Press, 112, pp. 207-225.
- Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 542, 553, 568.
- Uto, I., Ishimatsu, T., Hirayama, H., Ueda, S., Tsuruta, J. and Kambara, T. (1991). Determination of urinary Tamm-Horsfall protein by ELISA using a maleimide method for enzyme-antibody conjugation. *J. Immunol. Meth.* 138, 87-94.
- Iwai, K., Fukuoka, S.-I., Fushiki, T., Kido, K., Sengoku, Y. and Semba, T. (1988). Preparation of a verifiable peptide-protein immunogen: direction-controlled conjugation of a synthetic fragment of the monitor peptide with myoglobin and application for sequence analysis. *Anal. Biochem.* 171, 277-282.
- Ghetie, V., et al. (1990). *Bioconjugate Chem.* 1, 24-31.
- Carlsson, J., Drevin, H. and Axen, R. (1978). Protein thiolation and reversible protein-protein conjugation. N-succinimidyl-3-(2-pyridyldithio)propionate, a new heterobifunctional reagent. *Biochem. J.* 173, 723-737.
- Zarling, D.A., Watson, A. and Bach, F.H. (1980). Mapping of lymphocyte surface polypeptide antigens by chemical cross-linking with BSOCOES. *J. Immunol.* 124, 913-920.
- Park, L.S., Friend, D., Gillis, S. and Urdal, D.L. (1986). Characterization of the cell surface receptor for a multi-lineage colony-stimulating factor (CSF-2). *J. Biol. Chem.* 261, 205-210. (DST example)
- Browning, J. and Ribolini, A. (1989). Studies on the differing effects of tumor necrosis factor and lymphotoxin on the growth of several human tumor lines. *J. Immunol.* 143, 1859-1867. (EGS example)
- Myers, D.E., Uckun, F.M., Swaim, S.E. and Vallera, D.A. (1989). The effects of aromatic and aliphatic maleimide cross-linkers and anti-CD-5 ricin immuno-toxins. *J. Immunol. Meth.* 121, 129-142. (MBS example)

Pierce Cross-linking Reagent Literature References *continued*

43. Kitagawa, T. (1981). Enzyme labeling with *N*-hydroxysuccinimidyl ester of maleimide. *Enzyme Immunoassay*, Tokyo/New York, Igaku-Shoin pp. 81-89.
44. Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 266-268.
45. Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 239-242.
46. Samoszuk, M.K., Petersen, A., Lo-Hsueh, M. and Rietveld, C. (1989). A peroxide-generating immunoconjugate directed to eosinophil peroxidase is cytotoxic to Hodgkin's disease cells *in vitro*. *Antibody, Immunoconjugates and Radiopharmaceuticals* 2(1), 37-45.
47. Teale, J.M. and Kearney, J.R. (1986). Clonotypic analysis of the fetal B cell repertoire: Evidence for an early and predominant expression of idiotypes associated with the VH 36-60 family. *J. Mol. Cell. Immunol.* 2, 283-292.
48. Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 232-235.
49. Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 289, 291, 375.
50. *Previews*, May/June 1994, p. 1.
51. Lala, A., Sojar, H.T. and DeNardin, E. (1996). Quantitative approach for detection and characterization of formyl peptide receptor in solution using a photo affinity ligand. *Peptide Res.* 9(2), 58-60.
52. Bieniarz, C., Husain, M., Barnes, G., King, C.A. and Welch, C.J. (1996). Extended length heterobifunctional coupling agents for protein conjugations. *Bioconjugate Chem.* 7, 88-95.
53. Vanin, E.F. and Ji, T.H. (1981). Synthesis and application of cleavable photoactive heterobifunctional reagents. *Biochemistry* 20, 6754-6760.
54. Thevenin, B., *et al.* (1991). A novel reagent for functionally directed site-specific fluorescent labeling of proteins. *Biophys. J.* 59, 358a.
55. Eager, J.E. and Savige, W.E. (1963). Photolysis and photo-oxidation of amino acids and peptides - VI. A study of the initiation of disulfide interchange by light irradiation. *Photochem. Photobiol.* 2, 25-37.
56. Schnaar, R.L., Langer, B.G. and Brandley, B.K. (1985). Reversible covalent immobilization of ligands and proteins on polyacrylamide gels. *Anal. Biochem.* 151, 268-281.
57. May, J.M. (1989). Selective labeling of the erythrocyte hexose carrier with a maleimide derivative of glucosamine: Relationship of an exofacial sulfhydryl to carrier conformation and structure. *Biochemistry* 28, 1718-1725.
58. Sayre, L.M., Larson, D.L., Takemori, A.E. and Portoghese, P.S. (1984). Design and synthesis of naltrexone-derived affinity labels with nonequilibrium opioid agonist and antagonist activities. Evidence for the existence of different mu receptor subtypes in different tissues. *J. Med. Chem.* 27(10), 1325-35.
59. Stalteri, M.A. and Mather, S.J. (1995). A cross-linked monoclonal antibody fragment for improved tumor targeting. *Bioconjugate Chem.* 6, 179-186.
60. Yi, F., Denker, B.M. and Neer, E.J. (1991). Structural and functional studies of cross-linked G α protein subunits. *J. Biol. Chem.* 266(6), 3900-3906.
61. Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego, Academic Press, pp. 114-116, 294. (periodate cleavage of vic-diols)
62. O'Sullivan, M., *et al.* (1979). Comparison of two methods of preparing enzyme-antibody conjugates: Application of these conjugates to enzyme immunoassay. *Anal. Biochem.* 100, 100-108.
63. Rich, D., Gesellchen, P.D., Tong, D., Cheung, A. and Buckner, C.K. (1975).
 - Alkylating derivatives of amino acids and peptides. Synthesis of *N*-maleoylamino acids, [1-(*N*-maleoyl)glycyl]cysteinyloxytocin. Effects on vasopressin-stimulated water loss from isolated toad bladder. *J. Med. Chem.* 18, 1004-1010.
64. Moroder, L., *et al.* (1983). Immunoassays of peptide hormones and their chemical aspects. *Biopolymers* 22, 481-486.
65. Han, J.C. and Han, G.Y. (1994). A procedure for quantitative determination of tris(2-carboxyethyl)phosphine, an odorless reducing agent more stable and effective than dithiothreitol. *Anal. Biochem.* 220, 5-10.
66. Griffith, D.G., Partis, M.D., Sharp, R.N. and Beechey, R.B. (1981). *N*-Polymethylenecarboxymaleimides. A new class of probes for membrane sulfhydryl groups. *FEBS Lett.* 134, 261-263.
67. Trail, P.A., *et al.* (1993). Cure of xenografted human carcinomas by BR96-doxorubicin immunoconjugates. *Science* 261, 212-215.
68. Peeters, J.M., Hazendonk, T.G., Beuvery, E.C. and Tesser, G.I. (1989). Comparison of four bifunctional reagents for coupling peptides to protein and the effect of these three moieties on the immunogenicity of the conjugates. *J. Immunol. Methods* 120, 133-143.
69. Hermanson, G.T. (1996). *Bioconjugate Techniques*, San Diego: Academic Press, pp. 243-245. (EMCS use can be modeled after GMBs.)
70. Yoshitake, S., Yamada, Y., Ishikawa, E. and Masseyeff, R. (1979). Conjugation of glucose oxidase from *Aspergillus niger* and rabbit antibodies using *N*-(4-carboxycyclohexylmethyl)-maleimide. *Eur. J. Biochem.* 101, 395-399.
71. Annunziato, M.E., Patel, U.S., Ranade, M. and Palumbo, P.S. (1993). *p*-Maleimidophenyl isocyanate: A novel heterobifunctional linker for hydroxyl to thiol coupling. *Bioconjugate Chem.* 4, 212-218.
72. Duncan, R.J.S., Weston, P.D. and Wrigglesworth, R. (1983). A new reagent which may be used to introduce sulfhydryl groups into protein, and its use in the preparation of conjugates for immunoassay. *Anal. Biochem.* 132, 68-73.
73. Inman, J.K., Highet, P.F., Kolodny, N. and Robey, F.A. (1991). Synthesis of *N*-alpha-(*tert*-butoxycarbonyl)-*N*-epsilon-[*N*-(bromoacetyl)-beta-alanyl]-L-lysine. Its use in peptide synthesis for placing a bromoacetyl cross-linking function at any desired sequence position. *Bioconjugate Chem.* 2, 458-463.
74. Thorpe, P.E., *et al.* (1984). Blockade of the galactose-binding sites of ricin by its linkage to antibody. Specific cytotoxic effects of the conjugates. *Eur. J. Biochem.* 140, 63-71.
75. Rector, E.S., Schwenk, R.J., Tse, K.S. and Sehon, A.H. (1978). A method for the preparation of protein-protein conjugates of predetermined composition. *J. Immunol. Methods* 24, 321-336.
76. Zera, J.J., *et al.* (1991). A carbohydrate-directed heterobifunctional cross-linking reagent for the synthesis of immunoconjugates. *Anal. Biochem.* 194, 156-162.
77. Friden, P.M., *et al.* (1993). Blood-brain barrier penetration and *in-vivo* activity of an NGF conjugate. *Science* 259, 373-377.
78. Masri, M.S. and Friedman, M. (1988). Protein reactions with methyl and ethyl vinyl sulfones. *J. Protein Chem.* 7, 49-54.
79. Morpurgo, M., *et al.* (1996). Preparation and characterization of poly(ethylene glycol)vinyl sulfone. *Bioconjugate Chem.* 7, 363-368.
80. Ishii, Y. and Lehrer, S.S. (1986). Effects of the state of the succinimido-ring on the fluorescence and structural properties of pyrene maleimide-labeled alpha alpha-tropomyosin. *Biophys. J.* 50, 75-89.
81. Fuji, N., Akaji, K., Hayashi, Y. and Yajima, H. (1985). Studies on peptides. CXXV 3-(3-*p*-methoxybenzylthiopropionyl)-thiazolidine-2-thione and its analogs as reagents for the introduction of the mercapto group into peptides and proteins. *Chem. Pharm. Bull.* 33, 362-367.
82. Pandurangi, R.S., Lusiak, R., Desai, S. and Kuntz, R.R. (1998). Chemistry of bifunctional photoprobes: Part 4: Synthesis of the chromogenic, cleavable, wat soluble and heterobifunctional (*N*-Methyl amino perfluoroaryl azide benzamido ethyl-1,3-dithiopropionyl) sulfosuccinimide: An efficient protein cross-linking agent. *J. Chem. Soc. Chem. Commun.* Submitted for publication.
83. Pandurangi, R.S., Karra, R.S. and Volkert, W.A. (1997). Recent trends in the evaluation of the photochemical insertion characteristics of heterobifunctional perfluoroaryl azide chelating agents: Biochemical implication in nuclear medicine. *Photochem. Photobiol.* 65(2), 208-221.
84. Chrisey, L.A., Lee, G.U. and O'Ferrall, C.E. (1996). Covalent attachment of synthetic DNA to self-assembled monolayer films. *Nucleic Acids Research* 24(15) 3031-3039.
85. Rajur, S.B., Roth, C.M., Morgan, J.R. and Yarmush, M.L. (1997). Covalent protein-oligonucleotide conjugates for efficient delivery of antisense molecules. *Bioconjugate Chem.* 8, 935-940.
86. Wang, D., *et al.* (1997). Generation and characterization of an anti-CD19 single-chain Fv immunotoxin composed of C-terminal disulfide-linked dgRTA. *Bioconjugate Chem.* 8, 878-884.
87. Liver, D., *et al.* (1998). *Helicobacter pylori* adhesion binding fucosylated histo-blood group antigens revealed by retagging. *Science* 279, 373-376.
88. Grabarek, Z. and Gergely, J. (1990). Zero-length cross-linking procedure with the use of active esters. *Anal. Biochem.* 185(1), 131-135.
89. Kuijpers, W.H., Bos, E.S., Kaspersen, F.M., Veeneman, G.H. and van Boeckel, C.A. (1993). Specific recognition of antibody-oligonucleotide conjugates by radiolabeled antisense nucleotides: a novel approach for two step radio-immunotherapy of cancer. *Bioconjugate Chem.* 4(1), 94-102.
90. Nakagami, S., Matsunaga, H., Oka, N. and Yamane, A. (1991). Preparation of enzyme-conjugated DNA probe and application to the universal probe system. *Anal. Biochem.* 198(1), 75-79.
91. Geselowitz, D.A. and Neumann, R.D. (1995). Quantitation of triple-helix formation using a photo-cross-linkable arylazide/biotin/oligonucleotide conjugate. *Bioconjugate Chem.* 6(4), 502-506.

Quick Guide to Pierce Protein Modification Tools

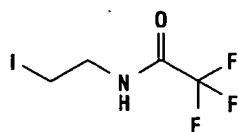
Primary Target of Reaction	Product #	Product Name	Use/Benefit
Disulfide groups	77700ZZ	Reduce-Imm™ Immobilized Reductant	Solid-phase system for reducing peptides and proteins
Disulfide groups	20290ZZ	Cleland's Reagent (DTT)	Conventional reducing agent for disulfide groups
Disulfide groups	35600ZZ	2-Mercaptoethanol	Mild reducing agent for cleaving disulfide bonds to thiols
Disulfide groups	20408ZZ	2-Mercaptoethylamine•HCl	Reduces disulfide groups; dissociates divalent IgG to monovalent IgG
Sulphydryl groups	33020ZZ	AMCA-HPDP	Used for fluorescent labeling*
Sulphydryl groups	26035ZZ	SBF-Chloride	Used for fluorescent detection
Sulphydryl groups	23010ZZ	Aminoethyl-8™ Reagent	Introduces primary amines
Sulphydryl groups	22582ZZ	Ellman's Reagent	Quantifies/detects sulphydryl groups in peptides and proteins
Sulphydryl groups	23030ZZ	N-Ethylmaleimide	Blocks sulphydryl groups
Sulphydryl groups	23012ZZ	2-Amino-2'-aminoethanethiolsulfonate	Converts sulphydryl groups to -S-S-CH ₂ CH ₂ NH ₂
Sulphydryl groups	22296ZZ	BMPA	Modifies sulphydryl groups to carboxyl groups
Sulphydryl groups	23011ZZ	MMTS	Converts sulphydryl groups on cysteine side chains into -S-S-CH ₃
Primary amines	20907ZZ	Citraconic Anhydride	Reversibly blocks primary amines
Primary amines	22333ZZ	DTPA	Introduces chelators for metal ions
Primary amines	27712ZZ	Water-Soluble Bolton-Hunter (Sulfo-SHPP)	Radioiodinates; introduces tyrosine groups**
Primary amines	27710ZZ	Bolton-Hunter (SHPP)	Radioiodinates; introduces tyrosine groups**
Primary amines	26777ZZ	Sulfo-NHS-Acetate	Blocks and acylates primary amines
Primary amines	26101ZZ	Traut's Reagent	Reacts with primary amines to introduce sulphydryl groups
Primary amines	26102ZZ	SATA	Reacts with primary amines to add protected sulphydryl groups
Primary amines	33005ZZ	AMCA-NHS	Used for fluorescent labeling*
Primary amines	33010ZZ	AMCA-Sulfo-NHS	Used for fluorescent labeling*
Thio esters	26103ZZ	Hydroxylamine•HCl	Deblocks SATA-modified proteins
Carbohydrates	33015ZZ	AMCA-Hydrazide	Used for fluorescent labeling*
Arginine	20100ZZ	p-Hydroxyphenylglyoxal	Detects arginine
Amines	28610ZZ	Sulfo-SDTB	Detects immobilized amino groups
Tryptophan residues	27580ZZ	BNPS-Skatole	Cleaves tryptophan residues

*Refer to page 230

**Refer to pages 232-233

Aminoethyl-8™ Reagent

Lets you modify free sulphydryls to primary amines.



Aminoethyl-8™ Reagent
M.W. 266.99

Features/Benefits:

- One-step modification reagent for sulphydryl groups
- At pH 8.1-8.6, the sulphydryl group is aminoethylated and the trifluoroacetyl group is hydrolyzed

Reference

1. Swartz, W.E., et al. (1980). *Anal. Biochem.* 106, 43-48.

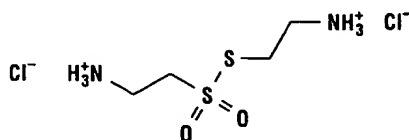
Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
23010ZZ	Aminoethyl-8™ Reagent (N-[Iodoethyl]trifluoroacetamide)	1 gm	\$ 73

800-874-3723

2-Aminoethyl-2'-aminoethanethiolsulfonate

Sulfhydryl-reactive amination reagent.



2-Aminoethyl-2'-aminoethanethiolsulfonate
M.W. 257.20
Spacer Arm 3.1 Å

Features/Benefits:

- 98% purity
- Converts sulfhydryl groups into -S-S-CH₂CH₂NH₂^{1,2}
- Cleavable with DTT or TCEP³
- Water-soluble

References

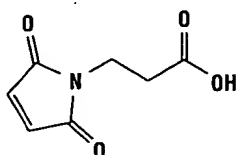
1. Field, L., Harle, H., Owen, T.C. and Ferretti, A. (1964). Preparation and oxidation of some asymmetrical dialkyl and alkyl pyridinium disulfides. *J. Org. Chem.* **29**, 1632-1635.
2. Field, L., Owen, T.C., Crenshaw, R.R. and Bryan, A.W. (1961). Thiosulfonates and disulfides containing 2-aminoethyl moieties. *J. Am. Chem. Soc.* **83**, 4414-4417.
3. Kirley, T.L. (1989). Reduction and fluorescent labeling of cyst(e)ine-containing proteins for subsequent structural analysis. *Anal. Biochem.* **180**, 231.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23012ZZ	2-Aminoethyl-2'-aminoethanethiolsulfonate	100 mg	\$ 2

BMPPA

Modifies sulfhydryl groups to carboxyl groups for the preparation of peptide-protein conjugates.



BMPPA
M.W. 169.13
Spacer Arm 5.9 Å

Features/Benefits:

- Sulfhydryl modification agent that creates a terminal -COOH group at -SH sites in proteins and other molecules

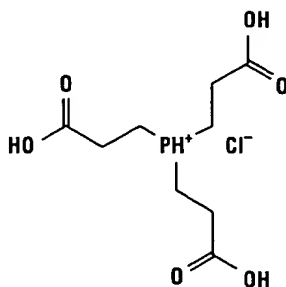
- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Makes -COOH groups available for coupling via EDC or other methods
- Non-cleavable

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22296ZZ	BMPPA (N-β-Maleimidopropionic acid)	100 mg	\$ 2

TCEP•HCl

Potent, water-soluble, odorless reducing agent in a conventional format.



TCEP•HCl
M.W. 286.65

Features/Benefits:

- Selective and complete reduction of even the most stable water-soluble alkyl disulfides

- Effective reduction at room temperature and pH 5 in less than five minutes
- Water solubility of 310 gm/L
- Resistant to air oxidation; nonvolatile and nonreactive toward other functional groups found in proteins

Reference

1. Kirley, T.L. (1989). *Anal. Biochem.* **180**, 231.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20490ZZ	TCEP•HCl (Tris[2-carboxyethyl]phosphine hydrochloride)	1 gm	\$

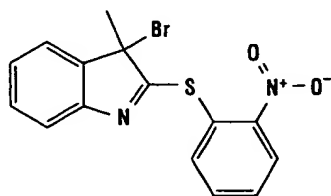
3

3-4

216

BNPS-Skatole

Useful for cleaving tryptophan residues.



BNPS-Skatole
M.W. 363.23

Reference

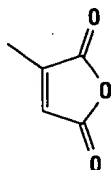
1. Fontana, A. (1972). *Methods in Enzymology* 25, 419-423.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27580ZZ	BNPS-Skatole (2-[2'-Nitrophenylsulfonyl]-3-methyl-3-bromoindolenine)	1 gm	\$137

Citraconic Anhydride

Reversibly blocks primary amines at pH 8.



Citraconic Anhydride
M.W. 112.08

Reference

1. Klapper, M.H. and Klotz, I.M. (1972). *Methods in Enzymology* 25, 531-552.

Ordering Information

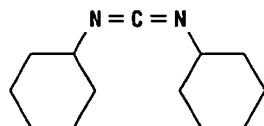
Product #	Description	Pkg. Size	U.S. Price
20907ZZ	Citraconic Anhydride	100 gm	\$ 50

Features/Benefits:

- Deblocking of lysine groups occurs under acidic conditions
- Modified enzymes have been reported to retain enzymatic activity after deblocking

DCC

One of the most frequently used coupling agents in peptide synthesis.



DCC
M.W. 206.33

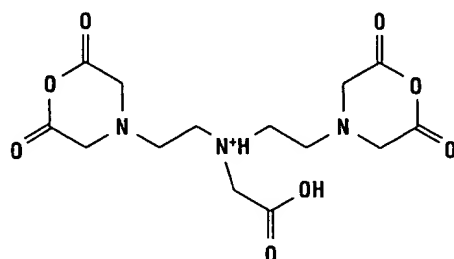
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20320ZZ	DCC (N,N'-Dicyclohexylcarbodiimide)	100 gm	\$ 19

800-874-3723

DTPA

Useful for rapid radiolabeling with Indium-III.



DTPA
M.W. 358.32

Features/Benefits:

- Used to introduce a strong chelator into proteins for rapid radiolabeling with Indium-III and other radionucleotides
- Will couple to antibodies at pH 7

Reference

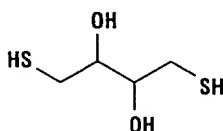
1. Layne, W.W., *et al.* (1982). *J. Nucl. Med.* 23, 627-630.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22333ZZ	DTPA (Diethylenetriaminepentaacetic anhydride)	1 gm	\$ 58

DTT

A water-soluble reagent that reduces disulfide bonds.



DTT
M.W. 154.25

Reference

1. Zahler, W.L. and Cleland, W.W. (1964). *Biochemistry* 3, 480-482.

Ordering Information

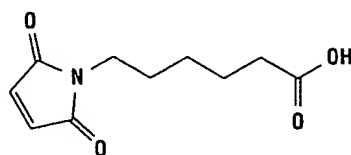
Product #	Description	Pkg. Size	U.S. Price
20290ZZ	DTT, Cleland's Reagent (Dithiothreitol)	5 gm	\$ 7

Compatible Product

22582ZZ	Ellman's Reagent (5,5'-Dithio-bis-[2-nitrobenzoic Acid])	5 gm	\$ 4
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EMCA

Useful for introducing maleimide groups into biomolecules.



EMCA
M.W. 211.21
Spacer Arm 9.4 Å

Features/Benefits:

- Sulfhydryl-reactive and amine-reactive via water-soluble carbodiimide (EDC) coupling

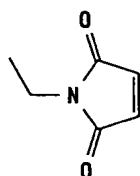
- Prepare maleimide-activated proteins through EDC coupling of the carboxyl group to available protein amino groups
- Maleimides react with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Modifies protein sulfhydryl groups to carboxyl groups
- Non-cleavable
- Probe for protein -SH groups

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22306ZZ	EMCA (N-ε-Maleimidocaproic acid)	1 gm	\$11

N-Ethylmaleimide

Useful for blocking free sulfhydryls.



N-Ethylmaleimide
M.W. 125.13

Features/Benefits:

- Forms covalent bonds with sulfhydryl groups at pH 6.5-7.5

- Reaction can be monitored by measuring the decrease in absorbance at 300 nm
- Has been used successfully for the blocking of sulfhydryl-containing reagents that interfere with enzyme assays

Reference

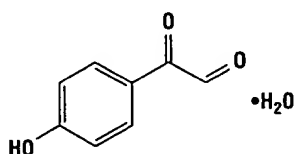
1. Partis, M.B., et al. (1983). *J. Prot. Chem.* 2, 263-277.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23030ZZ	N-Ethylmaleimide (NEM)	25 gm	\$ 85

HPG

An effective reagent for modifying arginine residues.



HPG
M.W. 168.15

Features/Benefits:

- Reacts specifically with arginine residues under mild conditions (pH 7-9, 25°C)

- Reaction follows Beer's Law at 5-50 μ M and can be monitored at 340 nm (pH 9)
- More resistant to oxidation than *p*-nitrophenylglyoxal and more water-soluble than phenylglyoxal

Reference

1. Yamasaki, R.B., et al. (1980). *Anal. Biochem.* 109, 32-40.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20100ZZ	HPG (<i>p</i> -Hydroxyphenylglyoxal)	100 mg	\$ 55

Hydroxylamine•HCl

A highly effective reagent for deblocking SATA-modified proteins and generating a free thiol.



Hydroxylamine Hydrochloride
M.W. 69.49

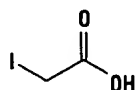
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26103ZZ	Hydroxylamine Hydrochloride	25 gm	\$ 28

• 800-874-3723

Iodoacetic Acid

For carboxymethylation.



Iodoacetic Acid
M.W. 185.95

Reference

1. Hall, J., et al. (1989). *Biochemistry* 28, 2568.

Ordering Information

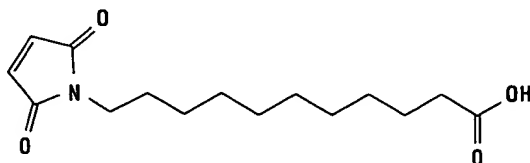
Product #	Description	Pkg. Size	U.S. Price
35603ZZ	Iodoacetic Acid	500 mg	\$ 34

2-Iminothiolane

See Traut's Reagent on page 225.

KMUA

Activates biomolecules for cross-linking through sulfhydryl groups or introduces carboxyl groups into proteins.



KMUA
M.W. 281.35
Spacer Arm 15.7 Å

Features/Benefits:

- Maleimide activate protein/peptide via EDC activation of the carboxyl group
- Sulfhydryl modification agent that creates a terminal carboxylate group at -SH sites in proteins and other molecules

- Maleimide reacts with -SH groups at pH 6.5-7.5, forming stable thioether linkages
- Non-cleavable, long aliphatic cross-bridge
- Useful for preparing peptide-protein conjugates

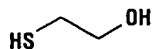
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22211ZZ	KMUA* (N- κ -Maleimidoundecanoic acid)	100 mg	\$ 2

*See also: BMPA, EMCA.

2-Mercaptoethanol

A mild reducing agent that is ideal for cleaving disulfide bonds to thiols.



2-Mercaptoethanol
M.W. 78.13

Features/Benefits:

- Also known as β -Mercaptoethanol
- Often included in enzyme solutions to protect against catalytic site inactivation due to cysteine sulfhydryl oxidation/disulfide formation; added at final concentrations of 5 and 20 mM, with or without EDTA as an additional protectant

Reference

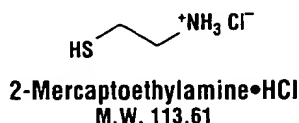
1. Yoshitake, S., et al. (1979). *Eur. J. Biochem.* 101, 395-399.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
35601ZZ	2-Mercaptoethanol	6 x 1 ml	\$ 2
35600ZZ	2-Mercaptoethanol	500 gm	\$ 3

2-Mercaptoethylamine•HCl

Dissociates divalent IgG to monovalent IgG without dissociating heavy and light chains.



Reference

1. Yoshitake, S., et al. (1979). *Eur. J. Biochem.* 101, 395-399.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20408ZZ	2-Mercaptoethylamine•HCl	6 x 6 mg	\$ 92

MMTS

Sulfhydryl-reactive and reversible sulfonating reagent.

Features/Benefits:

- 98% purity
- Converts sulfhydryl groups on cysteine side chains into -S-S-CH₃; free sulfhydryl is liberated with DTT or TCEP¹
- Used to modify thiol groups in creatine kinase²
- Boiling point: 85-87°C/0.8 mm

References

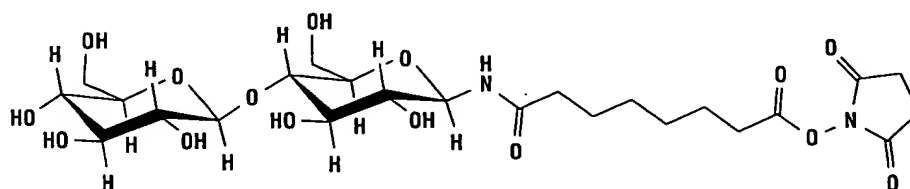
1. Kirley, T.L. (1989). Reduction and fluorescent labeling of cyst(e)ine-containing proteins for subsequent structural analyses. *Anal. Biochem.* 180, 231.
2. Smith, D.J., Maggio, E.T. and Kenyon, G.L. Simple alkanethiol groups for temporary blocking of enzymes. (1975). *Biochemistry* 14, 766.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23011ZZ	MMTS (Methyl methanethiosulfonate)	200 mg	\$ 28

Mono(lactosylamido) mono(succinimidyl)suberate

Amine-reactive lactosyl reagent.¹



Mono(lactosylamido) mono(succinimidyl)suberate
M.W. 594.56

Features/Benefits:

- For conjugation to amine-containing biomolecules or surfaces
- Useful in drug delivery and transport
- Imparts water solubility to conjugates

Reference

1. Vetter, D., Tate, E.M. and Gallop, M.A. (1995). Strategies for the synthesis and screening of glycoconjugates. Covalent immobilization for flow cytometry *Bioconjugate Chem.* 6, 319-322.

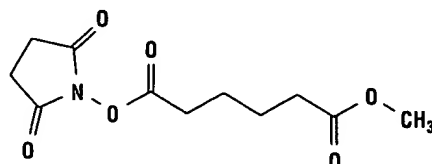
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23014ZZ	Mono(lactosylamido) mono(succinimidyl)suberate	50 mg	\$112

• 800-874-3723

MSA

Amine-reactive with latent carboxyl group.



MSA
M.W. 257.24
Spacer Arm 7.2 Å

Features/Benefits:

- Amine-reactive modification reagent containing a masked carboxyl group
- NHS ester reacts with primary amines at pH 7-9 to form stable amide bonds

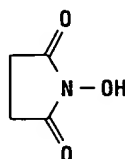
- Masked carboxyl group containing heterobifunctional reagent
- Converts amino groups to carboxyl groups
- Carboxyl group freed at pH 9.5 in phosphate buffer
- Non-cleavable; water-insoluble

Ordering Information

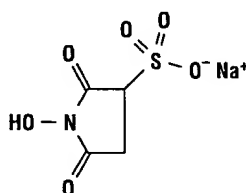
Product #	Description	Pkg. Size	U.S. Price
22605ZZ	MSA (Methyl <i>N</i> -succinimidyl adipate)	50 mg	\$ 45

NHS and Sulfo-NHS

Form amine-reactive peptides.



NHS
M.W. 115.09



Sulfo-NHS
M.W. 217.13

Feature/Benefit:

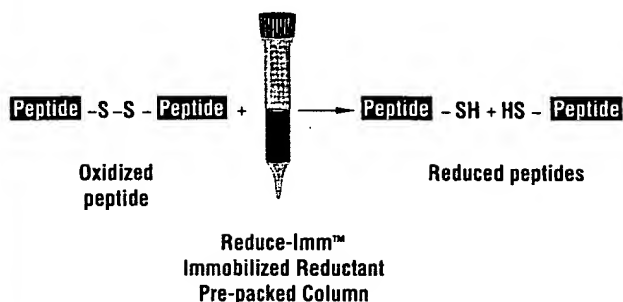
- COOH-containing molecule will form an acyl amino ester with NHS or Sulfo-NHS using a carbodiimide; this molecule will react with primary or secondary amines, yielding a stable amide bond

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
24500ZZ	NHS (<i>N</i> -Hydroxysuccinimide)	25 gm	\$ 20
24510ZZ	Sulfo-NHS (<i>N</i> -Hydroxysulfosuccinimide)	500 mg	\$ 98

Reduce-Imm™ Reducing Kit

Solid-phase system for reducing peptides and proteins.



Features/Benefits:

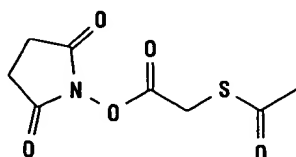
- Solid phase
- Excellent protein recovery
- Regenerable
- Protocols for both peptides and proteins

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
77700ZZ	Reduce-Imm™ Reducing Kit Sufficient reagents to reduce 150 µmoles of disulfide. Includes: Reduce-Imm™ Immobilized Reductant 2 x 2 ml Columns (capacity: 30-40 µmoles of sulfhydryl groups per column) Reduce-Imm™ Equilibration Buffer #1 Reduce-Imm™ Equilibration Buffer #2 DTT (used to regenerate and prime the column) Ellman's Reagent	Kit	\$299
77701ZZ	Reduce-Imm™ Immobilized Reductant Column	1 x 2 ml	\$109

SATA

Adds protected sulfhydryls to prevent disulfide formation.



SATA
M.W. 231.23
Spacer Arm 2.8 Å

Features/Benefits:

- Reacts with primary amines to add protected sulfhydryls
- When a free sulfhydryl is needed, an easy deprotection step is performed, generating a thioacetylated peptide; this free sulfhydryl-containing peptide can then be conjugated to form a hapten-carrier conjugate

Reference

1. Duncan, R.J.S., *et al.* (1983). *Anal. Biochem.* 132, 68-73.

Ordering Information

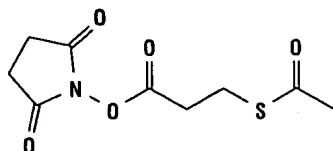
Product #	Description	Pkg. Size	U.S. Price
26102ZZ	SATA (N-Succinimidyl S-Acetylthioacetate)	50 mg	\$ 30

Compatible Products

20684ZZ	DMSO	50 ml	\$ 23
26103ZZ	Hydroxylamine Hydrochloride	25 gm	\$ 28

SATP

Same function as SATA, but offers more steric freedom for the unmasked sulfhydryl group.



SATP
M.W. 245.25
Spacer Arm 4.1 Å

Features/Benefits:

- Thiolation reagent that reacts with amines to add protected sulfhydryl groups
- Converts amino groups to sulfhydryl groups
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds

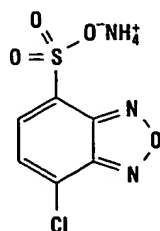
- Latent -SH group is released by hydroxylamine treatment and available for reaction with maleimide-activated biomolecules or other -SH group-containing compounds
- Cross-links formed with other -SH-containing molecules are reversible by reducing agents
- Modify peptides to facilitate the preparation of hapten-carrier conjugates

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26100ZZ	SATP (N-Succinimidyl S-acetylthiopropionate)	50 mg	\$ 49

SBF-Chloride

A water-soluble, nonmutagenic, thiol-specific, fluorescent probe.



SBF-Chloride
M.W. 251.65

Features/Benefits:

- An excellent replacement for 4-chloro-7-nitrobenzofurazan; SBF-Chloride has better solubility, specificity and is non-mutagenic

Reference

1. Andrews, J.L., *et al.* (1982). *Arch. Biochem. Biophys.* 214, 386-396.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26035ZZ	SBF-Chloride (Ammonium 4-chloro-7-sulfobenzofurazan)	100 mg	\$ 66

800-874-3723

Sodium meta-Periodate

An oxidation reagent of choice for creating active aldehydes for use in solid phase and solution coupling strategies.

Features/Benefits:

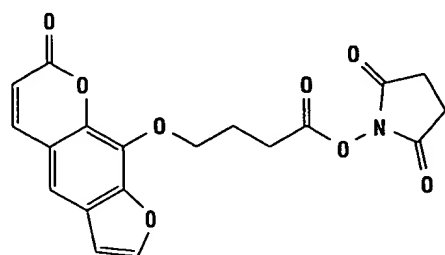
- White, crystalline, free-flowing
- A.C.S. reagent grade
- Capable of the oxidation of *cis*-diols to aldehydes

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20504ZZ	Sodium meta-Periodate	25 gm	\$ 18

SPB

Amine-reactive psoralen reagent.



SPB
M.W. 385.32

Features/Benefits:

- DNA intercalating agent and nucleic acid probe^{1,2}
- Reagent for photo-immobilization to microwell plates³
- Melting point: 177-178°C

References

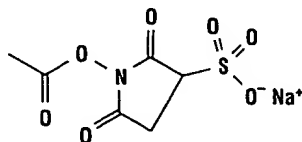
1. Laskin, J.D., *et al.* (1986). Psoralen potentiate ultraviolet light induced inhibition of epidermal growth factor binding. *Proc. Natl. Acad. Sci. USA* 83, 8211.
2. Inman, R.B. and Schnos, M. (1987). Electron microscopic identification of supercoiled regions in a complex DNA structure. *J. Mol. Biol.* 193, 377.
3. Elsner, H.I. and Mouritsen, S. (1994). Use of psoralens for covalent immobilization of biomolecules in solid phase assays. *Bioconjugate Chem.* 5, 463-467.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23013ZZ	SPB (Succinimidyl-[4-(psoralen-8-yloxy)]-butyrate)	50 mg	\$ 89

Sulfo-NHS Acetate

A water-soluble acylating reagent that blocks primary amines at pH 7 or greater.



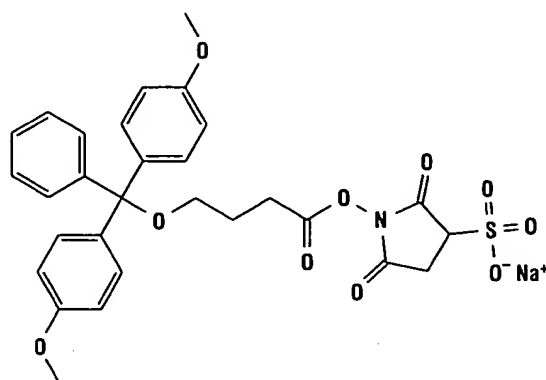
Sulfo-NHS Acetate
M.W. 259.17

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26777ZZ	Sulfo-NHS Acetate (Sulfosuccinimidyl Acetate)	100 mg	\$ 81

Sulfo-SDTB

Detects immobilized amino groups.



Sulfo-SDTB
M.W. 605.59

Features/Benefits:

- Useful for estimating the amine content on solid supports
- Sulfo-SDTB does not require a catalyst
- Treatment of a Sulfo-SDTB derivatized support results in the release of dimethoxytrityl cation, which is detected spectrophotometrically at 498 nm

References

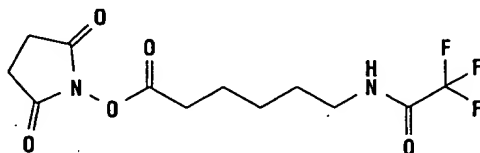
1. Gaur, R.K. and Gupta, K.C. (1989). *Anal. Biochem.* 180, 253-258.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28610ZZ	Sulfo-SDTB (Sulfosuccinimidyl-4-O-[4,4'-dimethoxytrityl]butyrate)	50 mg	\$107

TFCS

Amine-reactive cross-linker with a latent amino group available on demand.



TFCS
M.W. 324.25
Spacer Arm 7.7 Å

Features/Benefits:

- Amine-reactive modification agent with a protected primary amine group
- NHS ester end couples with primary amines at pH 7-9 to form stable amide bonds

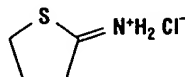
- Used to extend lysine side-chain length to reduce steric hindrance
- Temporarily block amine groups in target molecules
- Trifluoroacetyl protecting group released by phosphate or borate buffer, pH 7.8-8.1
- Unmasked amino group ready for reaction with any amine-reactive cross-linker for conjugate preparation
- Non-cleavable; water-insoluble

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22299ZZ	TFCS (N-[ε-Trifluoroacetylcaproyloxy]succinimide ester)	50 mg	\$ 39

Traut's Reagent

A water-soluble reagent that reacts with primary amines at pH 7-10 to introduce sulfhydryl groups.



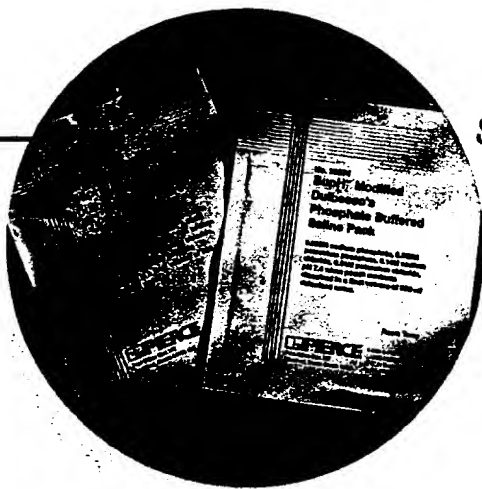
Traut's Reagent
M.W. 137.63
Spacer Arm 8.1 Å

Reference

1. Ghosh, S.S., et al. (1990). *Bioconjugate Chem.* 1, 71-76.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26101ZZ	Traut's Reagent (2-Iminothiolane-HCl)	500 mg	\$ 77



Special Product

BupH™ Dry Buffers

The most advanced, versatile, time-saving buffer product line available.

The ultimate in convenience ...

1. Reach for the sealed foil pack sitting conveniently on your bench top.
2. Open and add water.
3. The fresh buffer is ready-to-use in practical aliquots so there's no waste.

The ultimate in versatility ...

1. Routine buffers are designed for use in dialysis, cross-linking, enzyme assays, ELISAs, immunohistochemistry, protein plate-coating, biotinylation and other applications.
2. Using one buffer source maintains consistency and eliminates variables within the lab.
3. Specialized buffers ideally support your work in specific chemistries and methods.

The ultimate in integrity ...

1. Unlike stored buffers, BupH™ Buffers are protected from contamination.
2. Carry out applications with confidence in buffer quality.
3. "Test-assured" with the Pierce commitment to world-class, quality management standards.

The ultimate in timesavings ...

1. The making of specialized and routine buffers are no longer time-consuming.
2. No component measurement, pH adjustment, quality validation, preparation tracking or refrigeration hassle.
3. Move forward with your work by eliminating re-tests due to buffer problems.

BupH™ Borate Buffer Packs

Eliminate the need to pH – ideal for protein modification procedures that require an alkaline pH.

Ordering Information

Product #	Description	Pkg. Size	U. Pr
28384ZZ	BupH™ Borate Buffer Packs Each pack yields 50 mM borate, pH 8.5 after adding 500 ml of deionized water (20 liters total).	40 pack	\$

BupH™ Carbonate-Bicarbonate Buffer Packs

Ideal for microplate coating for RIA and EIA techniques.

Ordering Information

Product #	Description	Pkg. Size	U. Pr
28382ZZ	BupH™ Carbonate-Bicarbonate Buffer Packs Each pack yields 0.2 M Carbonate-Bicarbonate Buffer, pH 9.4 when dissolved in 500 ml distilled water (20 liters total).	40 pack	\$

Internet: <http://www.pierce.com>

BupH™ Citrate-Carbonate Buffer Packs, pH 9*Ideal for use with Pierce UltraLink™ Supports.*

One pack of BupH™ Citrate-Carbonate Buffer Pack, pH 9 dissolved in 100 ml of deionized water yields 0.6 M sodium citrate, 0.1 M sodium carbonate, pH 9. This buffer is perfect for azlactone coupling using UltraLink™ Biosupport Medium (Product # 53110ZZ).

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28388ZZ	BupH™ Citrate-Carbonate Buffer Packs, pH 9 Each pack yields 100 ml (1 liter total).	10 pack	\$ 40

*Also see BupH™ Citrate-MOPS.***BupH™ Citrate-MOPS Buffer Packs***Ideal for use with Pierce UltraLink™ Supports.*

BupH™ Citrate-MOPS Buffer is designed for use with Pierce UltraLink™ Biosupport Medium (Product # 53110ZZ). One pack of BupH™ Citrate-MOPS Buffer dissolved in 100 ml of deionized water yields 0.6 M sodium citrate, 0.1 M MOPS buffer, pH 7.5.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28386ZZ	BupH™ Citrate-MOPS Buffer Packs Each pack yields 100 ml (1 liter total).	10 pack	\$ 68

BupH™ MES Buffered Saline Packs*Ideal for use with carbodiimide coupling chemistries.*

BupH™ MES Buffered Saline Packs are designed for use with carbodiimide coupling chemistries. They can be used with EDC (Product # 22980ZZ) or Immobilized DADPA (Product # 20266ZZ) and EDC. One pack of BupH™ MES Buffered Saline dissolved in 500 ml of water yields 0.1 M MES (2-[N-Morpholino]ethanesulfonic acid), 0.9% NaCl, pH 4.7.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28390ZZ	BupH™ MES Buffered Saline Packs Each pack yields 500 ml (5 liters total).	10 pack	\$ 73

BupH™ Modified Dulbecco's PBS Packs*A ready-to-use PBS for immunoassays.***Ordering Information**

Product #	Description	Pkg. Size	U.S. Price
28374ZZ	BupH™ Modified Dulbecco's PBS Packs Each pack yields 500 ml of 0.008 M sodium phosphate, 0.002 M potassium phosphate, 0.14 M sodium chloride and 0.01 M potassium chloride, pH 7.4 when dissolved in 500 ml distilled water (20 liters total).	40 pack	\$ 60

BupH™ Phosphate Buffered Saline Packs*Its ionic strength makes it ideal for cross-linking and biotinylation.***Ordering Information**

Product #	Description	Pkg. Size	U.S. Price
28372ZZ	BupH™ Phosphate Buffered Saline Packs Each pack yields 500 ml of 0.1 M phosphate, 0.15 M NaCl, pH 7.2 when dissolved in 500 ml distilled water (20 liters total).	40 pack	\$ 83

800-874-3723

BupH™ Tris-Glycine and Tris Buffered Saline

Great for Western blots!

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28380ZZ	BupH™ Tris-Glycine Buffer Packs Each pack yields 500 ml of 25 mM Tris and 192 mM Glycine at a pH of approximately 8 when dissolved in 400 ml distilled water and 100 ml of methanol (20 liters total).	40 pack	\$ 86
28376ZZ	BupH™ Tris Buffered Saline Packs Each pack yields 500 ml of 25 mM Tris, 0.15 M NaCl, pH 7.2 when dissolved in 500 ml distilled water (20 liters total).	40 pack	\$ 83
28379ZZ	BupH™ Tris Buffered Saline Packs Each pack yields 500 ml of 25 mM Tris, 0.15 M NaCl, pH 7.6 when dissolved in 500 ml distilled water (20 liters total).	10 pack	\$ 39
Compatible Product			
28320ZZ	Surfact-Amps™ 20 (Ampules of 10% Tween®-20)	6 x 10 ml	\$ 58

Tris-Glycine-SDS Buffer Packs

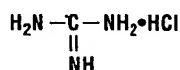
A ready-to-use electrophoresis buffer.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28378ZZ	BupH™ Tris-Glycine-SDS Buffer Packs Each pack yields 500 ml of 25 mM Tris, 192 mM Glycine and 0.1% SDS, pH 8.3 when dissolved in 500 ml distilled water (20 liters total).	40 pack	\$ 9

8 M Guanidine•HCl Solution and Guanidine•HCl

Ready-to-use, highly purified denaturants.



Guanidine Hydrochloride
M.W. 95.54

Features/Benefits:

- Free of UV-absorbing materials in the range of 225-300 nm
- Sharp UV cut-off spectrum with OD₂₆₀ less than 0.03
- Typical metals: Cu ≤ 1 ppm; Fe ≤ 0.1 ppm; Pb ≤ 0.1 ppm; Zn ≤ 0.1 ppm
- Particulate-free, crystal-clear, colorless solution
- Excellent stability
- Excellent for washing affinity ligand columns (nonprotein ligands)

8 M Guanidine•Hydrochloride Dilution Table

Beginning with 10 ml of Pierce 8 M Guanidine•HCl Solution (Product # 24115ZZ), dilution to the indicated final volume will give the stated molarity.

Molarity	Final Volume	Molarity	Final Volume
8 M	10 ml	3 M	26.7 ml
7 M	11.4 ml	2 M	40 ml
6 M	13.3 ml	1.5 M	52 ml
5 M	16 ml	1 M	80 ml
4 M	20 ml	0.5 M	160 ml

References

1. Tanaka, S., et al. (1985). *J. Biochem.* 97, 1377-1384.
2. Wong, K.P., et al. (1971). *Anal. Biochem.* 40, 459-464.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
24115ZZ	8 Molar Guanidine•HCl Solution Sequanal Grade	200 ml	\$11
24110ZZ	Guanidine•HCl Crystalline, Sequanal Grade	500 gm	\$11

Reacti-Bind™ DNA Coating Solution

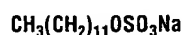
The fast and easy way to deposit target DNA onto a plastic surface.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
17250ZZ	Reacti-Bind™ DNA Coating Solution Sufficient for coating 5 x 96 well microwell plates (at 200 µl/well).	100 ml	\$ 35

SDS (Sodium Dodecyl Sulfate)

When resolution is the key, this is the ideal detergent.



SDS
M.W. 288.38

SDS (C₁₂)**Features/Benefits:**

- Greater than 99% alkyl sulfate
- Greater than 98% C₁₂ alkyl sulfate
- Contains a low level of hexadecyl sulfate C₁₆, which inhibits protein renaturation

SDS (Lauryl)**Features/Benefits:**

- Unique distribution of carbon chain lengths is advantageous when resolving viral proteins during gel electrophoresis
- Can be used for renaturation after SDS-PAGE (if gels are treated according to the procedure of Blank, *et al.* to remove C₁₄ and C₁₆ alkyl sulfates)

References

1. Matheka, H.D., *et al.* (1977). *Anal. Biochem.* 81, 9-17.
2. Swaney, J.B., *et al.* (1974). *Anal. Biochem.* 58, 337-346.
3. Blank, A., *et al.* (1980). *Federation Proceedings* 39(6), Abstracts ABSC/TBS, Abstract No. 1285, 1951.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28312ZZ	SDS, C ₁₂ Grade (Sodium Dodecyl Sulfate, C ₁₂)	500 gm	\$110
28364ZZ	SDS (Sodium Dodecyl Sulfate, lauryl)	100 gm	\$ 36
28365ZZ	SDS (Sodium Dodecyl Sulfate, lauryl) Typical Analysis: C ₁₂ : 63.5%, C ₁₄ : 29.5%, C ₁₆ : 7.0%	1 kg	\$135

Compatible Product

20346ZZ	Extracti-Gel™ Detergent Removing Gel AffinityPak™ Columns	5 x 1 ml	\$ 99
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Urea

A low UV-absorbing protein denaturant.

Features/Benefits:

- Melting point: 132-136°C
- Specification: A₂₈₀ < 0.100

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
29700ZZ	Urea, Sequanal Grade	1 kg	\$ 23

800-874-3723

Fluorescent Labels and Probes

The high sensitivity and resolution achieved with fluorescing compounds make fluorescent detection ideal for many immunological and biological techniques. Fluorescent labeling is widely used in immunohistochemistry, flow cytometry, fluorescence-activated cell sorter (FACS) analysis and molecular structure and function studies. Fluorescent labels are among the most sensitive for these applications, and they eliminate many of the problems associated with radiochemical labeling methods.

Many different fluorescent compounds are available for use in fluorescent labeling protocols. Each different fluorophore has distinct and characteristic excitation and emission spectra. Fluorochromes can be selected that have nonoverlapping emission spectra, and two or more fluorochromes can be visualized on the same sample.

Aldehyde/Ketone- and Cytidine-Reactive Probe

Product #	Probe Name	M.W.	Excitation Wavelength (nm)	Emission Wavelength (nm)	Comments	Pkg. Size	U.S. Price
33015ZZ	AMCA-Hydrazide	247.10	345	440-460	• Soluble in DMF	5 mg	\$ 91

Amine-Reactive Probes

Product #	Probe Name	M.W.	Excitation Wavelength (nm)	Emission Wavelength (nm)	Comments	Ref. #	Pkg. Size	U.S. Price
33005ZZ	AMCA-NHS	330.09	345	440-460			5 mg	\$ 71
33010ZZ	AMCA-Sulfo-NHS	431.02	345	440-460	• Water-soluble		5 mg	\$ 81
46117ZZ	DTAF•2HCl	568.20	492	510	• Superior stability	11,12	50 mg	\$ 41
46110ZZ	Fluorescein Isothiocyanate (FITC)	389	494	520	• Soluble in aqueous buffer > pH 6 and DMF	1-7	1 gm	\$131
46116ZZ	Fluorescein-5(6)-carboxamidoundecanoic acid-NHS-ester	659.69	492	510		13	25 mg	\$111
46100ZZ	NHS-Fluorescein	473	491	518	• Soluble in DMF and DMSO	6	100 mg	\$ 91
46102ZZ	NHS-Rhodamine	527	544	576	• Soluble in DMF and MeCN	6	25 mg	\$121
46118ZZ	Succinimidyl-acridine-9-carboxylate	320.30	364	460	• Blue fluorescent probe and oligonucleotide intercalating agent		50 mg	\$ 51
46112ZZ	Tetramethyl-rhodamine-5-(and 6)-isothiocyanate (TRITC)	479*	541	572	• Soluble in DMF	2,4	10 mg	\$161
46115ZZ	Texas Red® Sulfonyl Chloride	625	596	615	• Soluble in DMF and MeCN	2,9,10	10 mg	\$131

*Molecular weight based on the chloride salt

References

- Miki, M. and dos Remedios, C.G. (1988). Fluorescence quenching studies of fluorescein attached to lys-61 or cys-374 in actin: effects of polymerization, myosin sub fragment-1 binding, and tropomyosin-troponin binding. *J. Biochem.* **104**, 232-235.
- Smith, L.M., et al. (1987). The synthesis and use of fluorescent oligonucleotides in DNA sequence analysis. *Meth. Enzymol.* **155**, 260-301.
- Vera, J.C., et al. (1988). Purification, amino terminal analysis and peptide mapping of proteins after *in situ* postelectrophoretic fluorescent labeling. *Anal. Biochem.* **174**, 38-45.
- Szewczyk, B. and Summers, D.F. (1987). Fluorescent staining of proteins transferred to nitrocellulose allowing for subsequent probing with antisera. *Anal. Biochem.* **164**, 303-306.
- Der-Ballian, G.P., et al. (1988). Fluorescein labeling of Fab' while preserving single thiol. *Anal. Biochem.* **173**, 59-63.
- Vigers, G.P.A., et al. (1988). Fluorescent microtubules break up under illumination. *J. Cell Biol.* **107**, 1011.
- Goding, J.W. (1976). Conjugation of antibodies with fluorochrome: modifications to the standard methods. *J. Immunol. Methods* **13**, 215-226.
- Szewczyk, B., et al. (1987). Use of different fluorochromes for monitoring protein elution and transfer. *Electrophoresis* **8**, 25-28.
- Smith, L.M., et al. (1986). Fluorescence detection in automated DNA sequence analysis. *Nature* **321**, 674-678.
- Staines, W.A., et al. (1988). Three-color immunofluorescence histochemistry allowing triple labeling within a single section. *J. Histochem. Cytochem.* **36**(2) 145-151.
- Mahoney, C.W. and Azzi, A. (1987). The synthesis of fluorescent chlorotriaziny aminofluorescein-concanavalin A and its use as a glycoprotein stain on sodium dodecyl sulphate/polyacrylamide gels. *Biochem. J.* **243**(2), 569-574.
- Blakeslee, D. and Baines, M.G. (1976). Immunofluorescence using dichlorotriazinylaminofluorescein (DTAF). *J. Immunol. Methods* **13**, 305-320.
- Brinkley, M. (1992). A brief survey of methods for preparing protein conjugates with dyes, haptens, and cross-linking reagents. *Bioconjugate Chem.* **3**, 2.
- Reed, M.W., Adams, A.D., Nelson, J.S. and Meyer, R.B. (1991). Acridine- and cholesterol-derivatized solid supports for improved synthesis of 3'-modified oligonucleotides. *Bioconjugate Chem.* **2**, 217-225.
- Toulme, J.J., Kirsch, H.M., Loreau, N., Thuong, N.T. and Helene, C. (1986). Specific inhibition of mRNA translation by complementary oligonucleotides covalently linked to intercalating agents. *PNAS* **83**, 1227.

3

Internet: <http://www.pierce.com>

3-4

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Membrane Probe

Product #	Probe Name	M.W.	Excitation Wavelength (nm)	Emission Wavelength (nm)	Comments	Ref. #	Pkg. Size	U.S. Price
46164ZZ	1,6-Diphenyl-1,3,5-hexatriene (DPH)	232	348	426	• Soluble in MeCN and DMF* • Neutral membrane probe	1-2	1 gm	\$ 67

*DPH is frequently used from tetrahydrofuran solution, but this is not recommended because of possible peroxide formation of that solvent.

References

- Brasitus, T.A., et al. (1987). Dexamethasone-induced alterations in lipid composition and fluidity of rat proximal-small-intestinal brush-border membranes. *Biochem. J.* 248, 455-461.

- Adler, M. and Tritton, T.R. (1988). Fluorescence depolarization measurements on oriented membranes. *Biophys. J.* 53, 989-1005.

Phycobiliproteins

Product #	Probe Name	M.W.	Excitation Wavelength (nm)	Emission Wavelength (nm)	Ref. #	Pkg. Size	U.S. Price
46180ZZ	Allophycocyanin	104	650	660	1-4	2 mg	\$175
46185ZZ	R-Phycoerythrin	240	480, 545 and 565	578	1-6	2 mg	\$185

References

- Oi, V.T., et al. (1982). Fluorescent phycobiliprotein conjugates for analyses of cells and molecules. *J. Cell Biol.* 93, 981-986.
- Kronick, M.N. and Grossman, P.D. (1983). Immunoassay techniques with fluorescent phycobiliprotein conjugates. *Clin. Chem.* 29, 1582-1586.
- Parks, D.R. and Herzenberg, L.A. (1984). Fluorescence-activated cell sorting: theory, experimental optimization, and applications in lymphoid-cell biology. *Meth. Enzymol.* 108, 197-241.

- Kronick, M.N. (1986). The use of phycobiliproteins as fluorescent labels in immunoassay. *J. Immunol. Meth.* 92, 1-13.
- White, J.C. and Stryer, L. (1987). Photostability studies of phycobiliprotein fluorescent labels. *Anal. Biochem.* 161, 442-452.
- Festini, R., et al. (1987). Detection of triple antibody-binding lymphocytes in standard single laser flow cytometry using colloidal gold, fluorescein and phycoerythrin as labels. *J. Immunol. Meth.* 101, 23-28.

Site-Selective Probes

Product #	Probe Name	M.W.	Excitation Wavelength (nm)	Emission Wavelength (nm)	Comments	Ref. #	Pkg. Size	U.S. Price
46190ZZ	4',6-Diamidino-2-phenylindole hydrochloride (DAPI)	350	345	455	• Soluble in H ₂ O • Fluorescent retrograde tracer of neurons • Binds tightly to tubulin <i>in vitro</i> with no interference with microtubule assembly or GTP hydrolysis	1-4	10 mg	\$ 89
46170ZZ	Hoechst 33258 (bis-benzimide)	624	343	480	• Soluble in H ₂ O and DMF • Supravital minor groove-binding DNA stain	5-7	100 mg	\$ 36

References

- Kubista, M., et al. (1987). Characterization and interaction between DNA and 4',6-diamidino-2-phenylindole by optical spectroscopy. *Biochem.* 26, 4545-4553.
- Bonne, D., et al. (1985). 4',6-Diamidino-2-phenylindole, a fluorescent probe for tubulin and microtubules. *J. Biol. Chem.* 260(5), 2819-2825.
- Morikawa, K. and Yanagida, J. (1981). Visualization of individual DNA molecules in solution by light microscopy: DAPI staining method. *J. Biochem. (Tokyo)* 89, 693-696.
- Russell, W.C., et al. (1975). A simple cytochemical technique for demonstration of DNA in cells infected with mycoplasmas and viruses. *Nature* 253, 461-462.

- Mocharla, R., et al. (1987). A novel, sensitive fluorometric staining technique for the detection of DNA in RNA preparations. *Nucleic Acids Res.* 15(24), 10589.
- Downs, T.R. and Wilfinger, W.W. (1983). Fluorometric quantification of DNA in cells and tissue. *Anal. Biochem.* 131, 538-547.
- Latt, S.A. and Stetten, G. (1976). Spectral studies on 33258 Hoechst and related bisbenzimidazole dyes useful for fluorescent detection of deoxyribonucleic acid synthesis. *J. Histochem. Cytochem.* 24(1), 24-33.

Sulfhydryl-Reactive Probes

Product #	Probe Name	M.W.	Excitation Wavelength (nm)	Emission Wavelength (nm)	Comments	Ref. #	Pkg. Size	U.S. Price
33020ZZ	AMCA-HPDP	528.7	345	440-460			5 mg	\$ 91
46130ZZ	Fluorescein-5-maleimide	427	490	515	• Soluble in aqueous buffer > pH 6 and DMF	1	25 mg	\$129
46120ZZ	5-Iodoacetamido-fluorescein (5-IAF)	515	490	520	• Soluble in aqueous buffer > pH 6 and DMF	2-4	100 mg	\$201
46122ZZ	6-Iodoacetamido-fluorescein (6-IAF)	515	488	518	• Soluble in aqueous buffer > pH 6 and DMF	5,6	100 mg	\$201

References

- Curtis, S.K. and Cowden, R.R. (1980). Demonstration of sulfhydryl and disulfide groups by a fluorescent maleimide procedure. *Histochem.* 68, 23-28.
- Ansorge, W., et al. (1987). Automated DNA sequencing: ultrasensitive detection of fluorescent bands during electrophoresis. *Nucleic Acids Res.* 15(11), 4593-4602.
- Hartig, P.R., et al. (1977). 5-Iodoacetamidofluorescein-labeled chloroplast coupling factor 1: conformational dynamics and labeling-site characterization. *Biochem.* 16(19), 4275-4282.

- Bishop, J.E., et al. (1988). (Iodoacetamido)fluorescein labels a pair of proximal cysteines on the Ca²⁺ ATPase of sarcoplasmic reticulum. *Biochem.* 27(14), 5233-5240.
- Matteoni, R. and Kreis, T.E. (1987). Translocation and clustering of endosomes and lysosomes depends on microtubules. *J. Cell Biol.* 105, 1253-1265.
- Vandenbunder, B. and Borisy, G.G. (1986). Decoration of microtubules by fluorescently labeled microtubule-associated protein 2 (MAP2) does not interfere with their spatial organization and progress through mitosis in living fibroblasts. *Cell Mot. Cytoskel.* 6, 570-579.

Methods for Iodination of Proteins and Other Biomolecules

Radioiodination involves the introduction of radioactive iodine into certain amino acids (usually tyrosines) in proteins and peptides. Iodination takes place at the positions ortho to the hydroxyl group on tyrosine; mono- or di-substitution may occur. Studies on the mechanism of the reaction of iodine with tyrosines and other phenolic groups indicate that it is the phenolic anion that is attacked. Histidine residues are also iodinated by some iodinating methods. When iodlatable sites such as tyrosines are absent or of limited accessibility in a protein, iodlatable phenolic sites can be introduced by using the Bolton-Hunter Reagents (SHPP and Sulfo-SHPP).

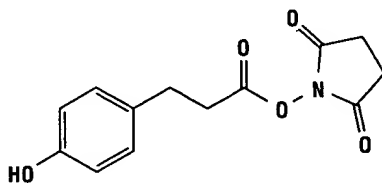
Radioactive ^{125}I or ^{131}I can be incorporated into proteins either by enzymatic or chemical oxidation. In the chemical oxidation method, Na^{125}I or Na^{131}I is converted to its corresponding reactive iodine form. For years, the oxidizing agent of choice for iodination was chloramine-T, which has strong oxidizing properties. The use of chloramine-T as an

iodination reagent, however, requires great care because its powerful oxidative properties may destroy the biological activity of a protein. After oxidation, the reaction is terminated by introduction of a reducing agent that also may affect the protein.

For maintaining the biological activity of proteins, the ideal oxidizing agent is one that is milder than chloramine-T, generates sufficient radioactive iodine and does not require a reduction step. Immobilization of the oxidant results in a two-phase system, limiting direct contact of the oxidant with the protein. A two-phase system allows for a slower and more easily-controlled reaction. Also, immobilization of an oxidation reagent allows for easy separation of the reagent from the reaction mixture. Three innovations in protein iodination, IODO-GEN® Pre-Coated Iodination Tubes, IODO-BEADS® Iodination Reagent and IODO-GEN® Iodination Reagent are described in this section.

Bolton-Hunter Reagent

Increases ^{125}I label on proteins.



**Bolton-Hunter Reagent
(SHPP)**
M.W. 263.25

Features/Benefits:

- Attaches tyrosine-like residues to primary amines to increase the yield of a subsequent iodination
- Reacts with *N*-terminal amino groups optimally at pH 8.5
- Iodinate before or after coupling to the molecule of interest

- Introduces tyrosyl moieties with a neutral linkage, through end-terminal α -amino groups or ϵ -amino groups of lysine
- Ideal for the iodination of proteins with no tyrosines, or tyrosines with limited accessibility to iodination
- Preserves tyrosines that might affect function or immunogenicity

Reference

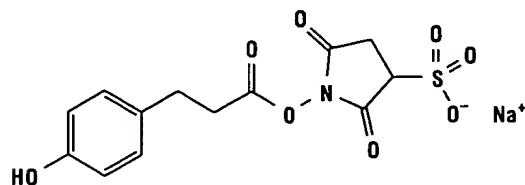
1. Bolton, A.E. and Hunter, W.M. (1973). The labeling of proteins to high specific radioactivities by conjugation to a ^{125}I -containing acylating agent. *Biochem. J.* 133, 529-539.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27710ZZ	Bolton-Hunter Reagent (SHPP) (<i>N</i> -Succinimidyl-3-[4-hydroxyphenyl]propionate)	1 gm	\$ 5

Water-Soluble Bolton Hunter Reagent

Membrane-impermeable and ideal for selectively radiolabeling cell surface proteins.



Water-Soluble Bolton-Hunter Reagent
(Sulfo-SHPP)
M.W. 365.29

Features/Benefits:

- Iodinate before or after coupling to the molecule of interest
- Useful for introducing tyrosyl groups on proteins sensitive to small amounts of organic solvent
- Useful for labeling cell surfaces without exposing the cells to membrane-permeable solvents^{1,2}

References

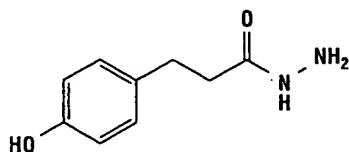
1. Thompson, J.A., Lau, A.L. and Cunningham, D.D. (1987). Selective radiolabeling of cell surface proteins to a high specific activity. *Biochem.* 26, 743-750.
2. Thirkell, D., Myles, A.D. and Russell, W.C. (1989). Serotype-8 and serocluster-specific surface-expressed antigens of *Ureaplasma urealyticum*. *Infection and Immunity* 57, 1697-1701.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27712ZZ	Water-Soluble Bolton-Hunter Reagent, Sulfo-SHPP (Sulfosuccinimidyl-3-[4-hydroxyphenyl]propionate)	100 mg	\$ 66

HPPH

Carbonyl- and aldehyde-reactive radioiodination reagent.



HPPH
M.W. 180.20
Spacer Arm 10.1 Å

Features/Benefits:

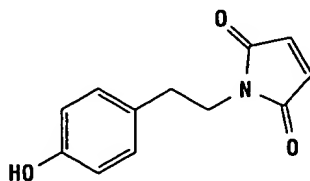
- 98% purity
- Hydrazide derivative of Bolton-Hunter Reagent
- White crystalline solid
- Melting point: 226°C

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27772ZZ	HPPH (3-[4-Hydroxyphenyl] propionic acid hydrazide)	100 mg	\$144

β-(4-Hydroxyphenyl)ethylmaleimide

Sulfhydryl-reactive radioiodination substrate.



β-(4-Hydroxyphenyl)ethyl maleimide
M.W. 217.22
Spacer Arm 10.1 Å

Features/Benefits:

- 97% purity
- Introduces 4-hydroxyphenyl ligand
- Readily radioiodinated
- Melting point: 136-137°C

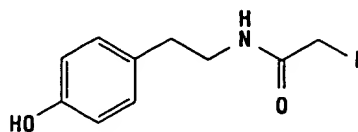
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27770ZZ	β-(4-Hydroxyphenyl)ethylmaleimide	100 mg	\$115

800-874-3723

β -(4-Hydroxyphenyl)ethyl iodoacetamide

Sulphydryl-reactive radioiodination substrate.



β -(4-Hydroxyphenyl)ethyl iodoacetamide

M.W. 305.11
Spacer Arm 10.1 Å

Features/Benefits:

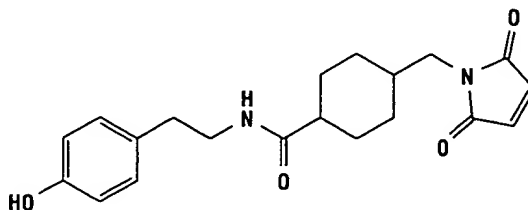
- 98% purity
- Introduces 4-hydroxyphenyl ligand
- Readily radioiodinated

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
27771ZZ	β -(4-Hydroxyphenyl)ethyl iodoacetamide	100 mg	\$11

2-(4-Hydroxyphenyl)ethyl 4-(N-maleimidomethyl)cyclohexane-1-carboxamide

Sulphydryl-reactive and extended radioiodination reagent.



2-(4-Hydroxyphenyl)ethyl 4-(N-maleimidomethyl)cyclohexane-1-carboxamide

M.W. 356.42
Spacer Arm 16.1 Å

Feature/Benefit:

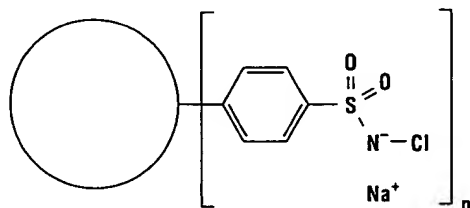
- 98% purity

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
27779ZZ	2-(4-Hydroxyphenyl)ethyl 4-(N-maleimidomethyl)-cyclohexane-1-carboxamide	100 mg	\$17

iodo-BEADS® Iodination Reagent

A convenient, gentle and effective method for iodinating soluble and membrane-bound proteins.

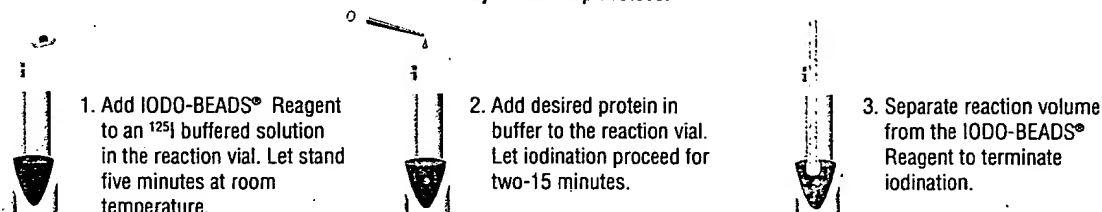


iodo-BEADS® Iodination Reagent

Features/Benefits:

- Derivatized, uniform, nonporous polystyrene beads
- Remarkably reproducible iodinations
- Radioiodide incorporation as high as 99%; labeled protein recovery > 90%
- Functions over a broad pH and temperature range
- Iodates in the presence of azides, detergents, urea and high salt
- Allows efficient iodination of cell membrane surface proteins
- More gentle method for iodination than soluble chloramine-T because there is no contact between the protein and the immobilized oxidizing agent¹
- Fast and easy to use ... iodinations complete in two to fifteen minutes
- Reaction stopped by simply removing beads from reaction mixture with tweezers or Pasteur pipet; no reducing agent necessary to terminate reaction
- More control over the incubation time
- Iodinate up to 500 µg of tyrosine-containing peptide or protein/bead
- Can be used to quantitatively iodinate histidine at pH 8.2²

Easy Three-Step Protocol



Recommended IODO-BEADS® Reaction Conditions

Protein or Peptide	5-500 µg of tyrosine-containing peptide or protein per bead
Beads	One or more; specific activity can be conveniently and reproducibly controlled by changing the number of beads
Reaction Volume	100-1,000 µl per bead; smaller volumes are possible using polypropylene Eppendorf tubes
Iodination Buffer	100 mM phosphate or Tris; IODO-BEADS® Iodination Reagent functions effectively in the presence of enzyme inhibitors such as azide, detergents, urea and high salt concentrations. Organic solvents that readily dissolve the polystyrene (such as DMSO or DMF) are incompatible with the use of the beads
pH	5.5-7.0; IODO-BEADS® Reagent functions best at pH 6.5 (pH 8.5 can be used, but with reduced iodination)
Temperature	iodo-BEADS® Iodination Reagent functions over a wide range of temperatures (even as low as 4°C), making it ideal for labile proteins
Time	Two-15 minutes; specific activity can be conveniently and reproducibly controlled by increasing or decreasing the incubation time

References

1. Markwell, M.A.K. (1982). A new solid-state reagent to iodinate proteins: conditions for the efficient labeling of antiserum. *Anal. Biochem.* 125, 427-432.
2. Tsomides, T.J., Walker, B.D. and Eisen, H.N. (1991). An optimal viral peptide recognized by CD8⁺ T cells binds very tightly to the restricting class I major histocompatibility complex protein on intact cells but not to the purified class I protein. *PNAS* 88, 11276-11280.

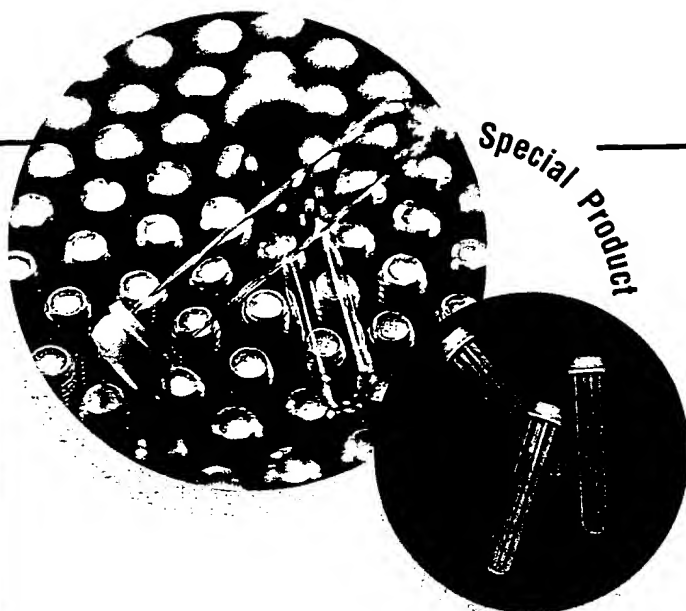
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28665ZZ	iodo-BEADS® Iodination Reagent*	50 beads	\$ 66
28666ZZ	iodo-BEADS® Iodination Reagent* (N-Chloro-benzenesulfonamide) Bead Diameter: 1/8 inch (3.175 mm), non-porous polystyrene Oxidative Capacity: 0.55 ± 0.05 µmoles/bead	250 beads	\$155

*U.S. Patent #'s 4,448,764 and 4,436,718

Iodinatable Reagent	Iodinate Prior to Coupling With:	Iodinate After Coupling With:
SHPP/Sulfo-SHPP	iodo-BEADS® Iodination Reagent or Chloramine-T	iodo-BEADS® Iodination Reagent or iodo-GEN® Iodination Reagent

Special Product



IODO-GEN® Pre-Coated Iodination Tubes

Unparalleled flexibility and reproducibility for radioiodinations.

The popularity of Pierce IODO-GEN® Iodination Reagent stems from its gentle and highly efficient chemistry.¹ Now Pierce is offering this reagent in the most convenient and versatile form ever ... pre-coating the inside surface of 12 mm x 75 mm test tubes.

Features/Benefits:

- Eliminates the tedious reagent surface coating step
- Produces a consistently IODO-GEN® Reagent-coated and flake-resistant surface
- Generates more than sufficient oxidative capacity for a typical protocol
- Allows iodinations to be carried out directly in the tube
- Enables ¹²⁵I to be pre-activated directly in the tube
- Offers the opportunity to iodinate protein without ever having the protein contact the IODO-GEN® Reagent directly

New ¹²⁵I pre-activation strategy² offers significant benefits, including:

- Elimination of oxidative damage to labile proteins
- No losses from nonspecific protein binding to the IODO-GEN® Reagent-coated surface
- Flexibility to conduct iodinations in a wide variety of vessels (microfuge tubes, tissue culture flasks, silanized tubes, etc.)
- Easy scale-up or scale-down of the iodination protocol
- Provides a very gentle environment to facilitate ¹²⁵I labeling of a wide range of proteins, peptides or cell surfaces
- Compatibility with common detergents

References

1. Fraker, P.J. and Speck, J.C. (1978). Protein and cell membrane iodinations with a sparingly soluble chloramide, 1,3,4,6-tetrachloro-3 α -6 α -diphenylglycoluril. *Biophys. Res. Comm.* **80**, 849-857.
2. Chizzonite, R. (1996). Hoffmann LaRoche, Department of Inflammation/Auto-immune Diseases, E-mail contact: Richard.Chizzonite@Roche.com.

Ordering Information

Product #	Description	Pkg. Size	U.S. Pric
28601ZZ	IODO-GEN® Pre-Coated Iodination Tubes Contains: 50 μ g IODO-GEN® Iodination Reagent evaporated from 100 μ l volume in 12 mm x 75 mm glass test tubes	10/pkg.	\$ 4!

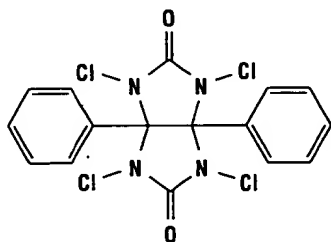
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3-4

236

ODO-GEN® Iodination Reagent

A solid-phase iodinating reagent ... for gentle iodinations.



ODO-GEN® Iodination Reagent
M.W. 432.09

Features/Benefits:

- Extremely stable and virtually insoluble in water¹
- Solubility in chloroform permits "plating" aliquots of the reagent onto the walls of the glass or plastic iodination vessels; plated vessels can be prepared in advance and stored in a dessicator
- Allows rapid iodination of proteins in an aqueous reaction mixture containing radioactive Na¹²⁵I
- Side reactions are negligible
- No reducing reagent is required because reactions are terminated by simply decanting the reaction solution away from the plated IODO-GEN® Iodination Reagent
- Specific activities of 1×10^5 cpm of ¹²⁵I per microgram of protein and labeling efficiencies of 60% are easily achieved
- Can be used to iodinate phenolic groups on cross-linkers or other protein modification reagents either before, or after, a protein, peptide or cell surface is reacted with the modification reagent
- Small amounts of protein can be iodinated²
- More efficient than chloramine-T or lactoperoxidase⁴
- Iodinate phenolic groups on cross-linking reagents (see list of compatible cross-linkers)

Applications:

- Recommended for use with ¹²⁵I³
- Substitutes effectively for the highly toxic TiCl₃ method for iodination of nucleic acids⁵
- Oxidatively remove sulfhydryl groups from solution prior to protein assay⁶

References

1. Fraker, P.J. and Speck, J.C., Jr. (1978). Protein and cell membrane iodinations with a sparingly soluble chloroamide, 1,3,4,6-tetrachloro-3α,6α-diphenylglycoluril. *Biochem. Biophys. Res. Comm.* **80**(4), 849-857.
2. Tolan, D.R., et al. (1980). Radioiodination of microgram quantities of ribosomal proteins from polyacrylamide gels. *Anal. Biochem.* **103**, 101-109.
3. Millar, W.T. and Smith, J.F.B. (1983). Protein iodination using IODO-GEN®. *Int. J. Appl. Radiat. Isot.* **34**(3), 639-641.
4. Salacinski, P.R.P., et al. (1981). Iodination of proteins, glycoproteins, and peptides using a solid-phase oxidizing agent, 1,3,4,6-tetrachloro-3α,6α-diphenylglycoluril (ODO-GEN®). *Anal. Biochem.* **117**, 136-146.
5. Piatyszek, M.A., Jarmolowski, A. and Augustyniak, J. (1988). IODO-GEN® mediated radioiodination of nucleic acids. *Anal. Biochem.* **172**, 356-359.
6. McClard, R.W. (1981). Removal of sulfhydryl groups with 1,3,4,6-tetrachloro-3α,6α-diphenylglycoluril: application to the assay of protein in the presence of thiol reagents. *Anal. Biochem.* **112**, 278-281.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
28600ZZ	ODO-GEN® Iodination Reagent (1,3,4,6-Tetrachloro-3α,6α-diphenylglycoluril)	1 gm	\$ 95

Compatible Cross-linkers (easily iodinated with IODO-GEN® Iodination Reagent)

27716ZZ	SASD	50 mg	\$161
27720ZZ	APDP	50 mg	\$199
27735ZZ	Sulfo-NHS-LC-ASA	50 mg	\$225
21564ZZ	BASED	50 mg	\$199
21512ZZ	ASBA	50 mg	\$ 95

Comparison of IODO-BEADS® and IODO-GEN® Iodination Reagents

	ODO-BEADS® Reagent	ODO-GEN® Pre-Coated Iodination Tubes	ODO-GEN® Reagent
Detergent- Compatible	Yes	Yes	Yes
Denaturant- Compatible	Yes	Yes	Yes
pH Range	4-8.5 (5-6.5 optimum)	4.4-9 (8-9 optimum)	4.4-9 (8-9 optimum)
Stability	Short - one year	Stable indefinitely	Stable indefinitely
Ease of Use	Easy	Easy	Requires good plating technique

3

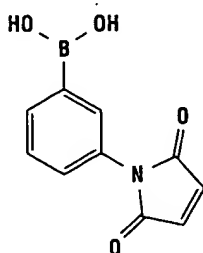
Protein Modification

3-4
237

800-874-3723

MPBA

Sulfhydryl-reactive radioiodination reagent.



MPBA
M.W. 216.99
Spacer Arm 8.8 Å

Features/Benefits:

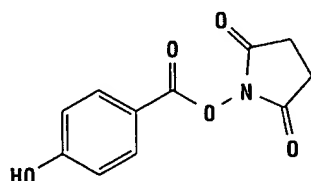
- 98% purity
- Melting point: > 360°C

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27778ZZ	MPBA (3-Maleimidophenyl boronic acid)	100 mg	\$ 86

SHB

Short, amine-reactive radioiodination reagent.



SHB
M.W. 235.19
Spacer Arm 5.7 Å

Features/Benefits:

- 97% purity
- Melting point: 175-176°C

Reference

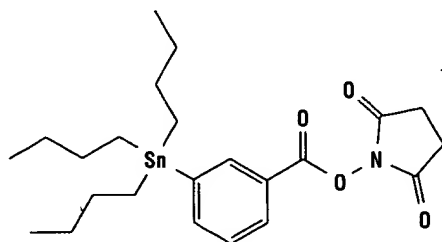
1. Vaidyanathan, G., Affleck, D.J. and Zalutsky, M.R. (1993). Radioiodination of proteins using *N*-succinimidyl 4-hydroxy-3-iodobenzoate. *Bioconjugate* 4, 78-84.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27774ZZ	SHB (Succinimidyl-4-hydroxybenzoate)	100 mg	\$ 69

Succinimidyl 3-(tri-*N*-butylstannyl)benzoate

Reacts with Na¹²⁵I to produce (SIB) succinimidyl iodobenzoate.



Succinimidyl 3-(tri-*N*-butylstannyl)benzoate
M.W. 508.24

Features/Benefits:

- 97% purity
- Colorless oil

References

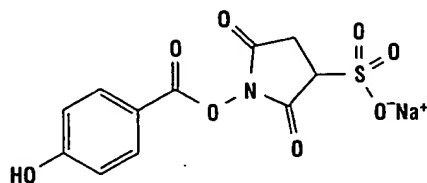
1. Zalutsky, M.R. and Narula, A.S. (1987). A method for the radiohalogenation of proteins resulting in decreased thyroid uptake of radioiodine. *Appl. Radiat. Isot.* 38, 1051-1055.
2. Garg, P.K., Alston, K.L. and Zalutsky, M.R. (1995). Catabolism of radioiodinated murine monoclonal antibody F(ab')₂ fragment labeled using *N*-succinimidyl 3-iodobenzoate and IODO-GEN® methods. *Bioconjugate Chem.* 6, 493-501.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27776ZZ	Succinimidyl 3-(tri- <i>N</i> -butylstannyl)benzoate	100 mg	\$111

Sulfo-SHB

Short, amine-reactive radioiodination reagent.



Sulfo-SHB
M.W. 337.24
Spacer Arm 5.7 Å

Feature/Benefit:

- Water-soluble analog of SHB¹

Reference

1. Vaidyanathan, G., Affleck, D.J. and Zalatsky, M.R. (1993). Radioiodination of proteins using *N*-succinimidyl 4-hydroxy-3-iodobenzoate. *Bioconjugate* 4, 78-84.

Ordering Information

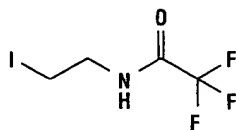
Product #	Description	Pkg. Size	U.S. Price
27775ZZ	Sulfo-SHB (Succinimidyl-4-hydroxybenzoate)	100 mg	\$ 86

• Chemical Cleavage Reagents

This section includes the reagents that chemically cleave proteins, or that are used to direct the chemical cleavage of proteins to specific sites.

Aminoethyl-8™ Reagent

A stable non-polymerizing substitute for ethylene imine.



Aminoethyl-8™ Reagent
M.W. 266.99

Features/Benefits:

- Aminoethyl-8™ Reagent is a one-step modification agent for the sulfhydryl group of cysteine

- The trypsin susceptible derivative, S-(β-aminoethyl)cysteine, results from the reaction
- Under the conditions of the alkylation reaction, the -SH group is ethylated and the trifluoroacetyl group is lost

Reference

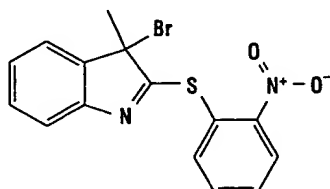
1. Schwartz, W.E., et al. (1980). *Anal. Biochem.* 106, 43-48.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23010ZZ	Aminoethyl-8™ Reagent (<i>N</i> -[Iodoethyl]trifluoroacetamide)	1 gm	\$ 73

BNPS-Skatole

Cleaves tryptophanyl peptides and proteins if sulfhydryl groups are protected.



BNPS-Skatole
M.W. 363.23

References

1. Fontana, A. (1972). *Methods in Enzymology* XXV, 419-423.
2. Fontana, A., et al. (1980). *Methods in Peptide and Protein Sequence Analysis*, Birr, Chr., Ed., Amsterdam: Elsevier, pp. 309-322.
3. Hunziker, P.E., et al. (1980). *Biochem. J.* 187, 515-519.

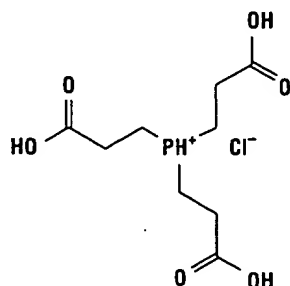
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27580ZZ	BNPS-Skatole (2-[2'-Nitrophenylsulfenyl]-3-methyl-3'-bromoindolenine)	1 gm	\$137

800-874-3723

TCEP HCl

Potent, water-soluble, odorless reducing agent in a conventional solid format.



TCEP•HCl
M.W. 286.65

Features/Benefits:

- Selective and complete reduction of even the most stable water-soluble alkyl disulfides

- Effective reduction at room temperature and pH 5 in less than five minutes
- Water solubility of 310 gm/L
- Resistant to air oxidation; nonvolatile and nonreactive toward other functional groups found in proteins

Reference

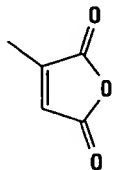
1. Kirley, T.L. (1989). *Anal. Biochem.* 180, 231.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20490ZZ	TCEP•HCl (Tris[2-carboxyethyl]phosphine hydrochloride)	1 gm	\$ 34

Citraconic Anhydride

Ideal for reversible primary amine blockage.



Citraconic Anhydride
M.W. 112.08

Features/Benefits:

- Reversibly blocks primary amines (epsilon amine group of lysine) at pH 8
- Deblocking of lysine groups occurs under acidic conditions
- Modified enzymes retain enzymatic activity after deblocking

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20907ZZ	Citraconic Anhydride	100 gm	\$ 51

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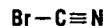
Internet: <http://www.pierce.com>

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Cyanogen Bromide

Selectively attacks methionine in proteins – an outstanding example of specific nonenzymatic cleavage.



Cyanogen Bromide
M.W. 105.9

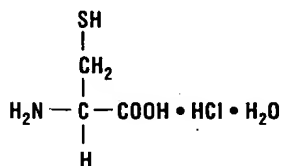
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
92278ZZ	Cyanogen Bromide	100 gm	\$ 57

Note: Federal regulations require that Cyanogen Bromide be shipped by motor freight only. There is an additional charge for shipping this item.

Cysteine•HCl

Useful as a sulfhydryl group standard.



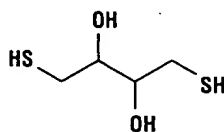
Cysteine•HCl•H₂O
M.W. 175.6

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
44889ZZ	Cysteine•HCl•H ₂ O	5 gm	\$ 33

DTT

A water-soluble reagent that reduces disulfide bonds.



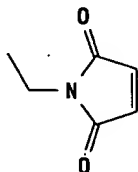
DTT
M.W. 154.25

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20290ZZ	DTT, Cleland's Reagent (Dithiothreitol)	5 gm	\$ 73

N-Ethylmaleimide

Useful for blocking free sulfhydryls.



N-Ethylmaleimide
M.W. 125.13

Features/Benefits:

- Forms covalent bonds with sulfhydryl groups at pH 6.5-7.5
- Reaction of the maleimide group with sulfhydryls can be monitored by measuring the decrease in absorbance at 300 nm

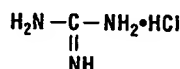
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23030ZZ	N-Ethylmaleimide (NEM)	25 gm	\$ 85

800-874-3723

Guanidine HCl

A highly purified denaturant.



Guanidine Hydrochloride
M.W. 95.54

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
24110ZZ	Guanidine•HCl Crystalline, Sequanal Grade	500 gm	\$121

8 M Guanidine•HCl Solution

Ready-to-use and easily diluted to any molarity below 8 M.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
24115ZZ	8 Molar Guanidine•HCl Solution Sequanal Grade	200 ml	\$101

Heptafluorobutyric Acid

An ion pair reagent for the reverse-phase HPLC separation of proteins and peptides.



Heptafluorobutyric Acid
M.W. 214.04

Features/Benefits:

- Clear, colorless liquid
- Typical purity is 99.7% by GC; less than 0.1% water

References

1. Hearn, M.T.W. and Hancock, W.S. (1979). *Trends in Biochemical Sciences* 4, N58-N62.
2. Bennett, H.P.J., et al. (1980). *J. Liquid Chrom.* 3, 1353-1365.
3. Bennett, H.P.J., et al. (1981). *Biochem.* 20, 4530-4538.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
25003ZZ	Heptafluorobutyric Acid Purity: ≥ 99.5% Density: 1.645 Boiling Point: 120°C	100 ml	\$241

Hydrochloric Acid in Dioxane

Ready-to-use for t-Boc group removal.

Features/Benefits:

- Reduces loss of synthetic peptide from support
- No cleavage of peptide-to-resin ester linkage
- Quantitative deprotection

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
24312ZZ	4N Hydrochloric Acid in Dioxane Sequanal Grade; low peroxide dioxane is gassed with anhydrous HCl to a normality of 4.05 ± 0.05. Sealed in glass ampules under nitrogen.	12 x 10 ml	\$173

Also Available:

24309ZZ	6N Hydrochloric Acid	25 x 1 ml	\$ 58
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Hydroxylamine HCl

It deblocks SATA-modified proteins and cleaves Asn-Gly bonds.



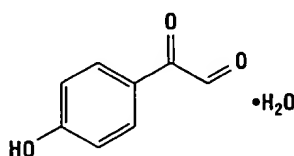
Hydroxylamine Hydrochloride
M.W. 69.49

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
26103ZZ	Hydroxylamine•HCl	25 gm	\$ 28

HPG (*p*-Hydroxyphenylglyoxal)

An effective reagent for modifying arginine residues.



HPG
M.W. 168.15

Features/Benefits:

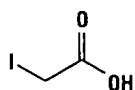
- Reacts specifically with arginine residues under mild conditions (pH 7-9; 25°C)
- Reaction follows Beer's Law at 5-50 μM and can be monitored at 340 nm (pH 9)
- More resistant to oxidation than *p*-nitrophenylglyoxal and more water-soluble than phenylglyoxal

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
20100ZZ	HPG (<i>p</i> -Hydroxyphenylglyoxal)	100 mg	\$ 55

Iodoacetic Acid

For carboxymethylation of lysine and histidine.



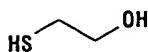
Iodoacetic Acid
M.W. 185.95

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
35603ZZ	Iodoacetic Acid	500 mg	\$ 34

2-Mercaptoethanol

A mild reducing agent for cleaving disulfide bonds to thiols.



2-Mercaptoethanol
M.W. 78.13

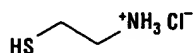
Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
35601ZZ	2-Mercaptoethanol	6 x 1 ml	\$ 27
35600ZZ	2-Mercaptoethanol	500 gm	\$ 33

800-874-3723

2-Mercaptoethylamine HCl

Dissociates divalent IgG to monovalent IgG without dissociating heavy and light chains.



2-Mercaptoethylamine•HCl
M.W. 113.61

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20408ZZ	2-Mercaptoethylamine•HCl	6 x 6 mg	\$ 1

Propionic Acid/Hydrochloric Acid

Analyzes the progress of peptide synthesis on a solid support without prior cleavage of the peptide from the resin.

Feature/Benefit:

- Hydrolysis is carried out at 130°C for two hours

Reference

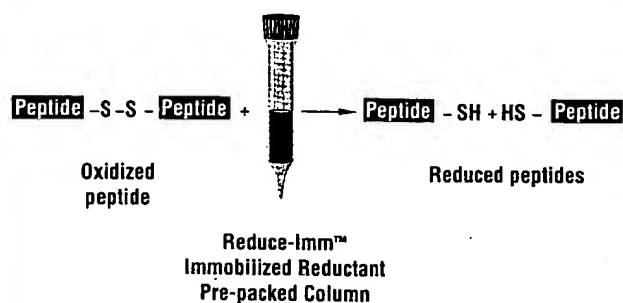
1. Westall, F.C., et al. (1972). *J. Org. Chem.* 37, 3363-3365.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
27514ZZ	Propionic Acid/Hydrochloric Acid 50:50 v/v, Sequanal Grade	25 x 1 ml	\$ 1

Reduce-Imm™ Reducing Kit

Solid-phase system for reducing peptides and proteins.

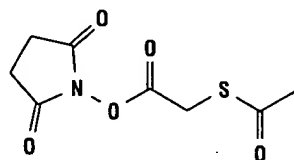


Ordering Information

Product #	Description	Pkg. Size	U.S. Price
77700ZZ	Reduce-Imm™ Reducing Kit Sufficient reagents to reduce 150 μmoles of disulfide.	Kit	\$21
77701ZZ	Reduce-Imm™ Immobilized Reductant Column	1 x 2 ml	\$11

SATA

Adds protected sulfhydryls to prevent dipeptide formation.



SATA
M.W. 231.23
Spacer Arm 2.8 Å

Reference

1. Duncan, R.J.S., Weston, P.D. and Wrigglesworth, R. (1983). A new reagent which may be used to introduce sulfhydryl groups into proteins, and its use in the preparation of conjugates for immunoassay. *Anal. Biochem.* 132, 68-73.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26102ZZ	SATA (N-Succinimidyl S-acetylthioacetate)	50 mg	\$ 1

Compatible Products

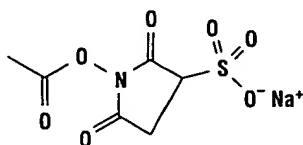
20684ZZ	DMSO	50 ml	\$ 1
26103ZZ	Hydroxylamine Hydrochloride	25 gm	\$ 1

Features/Benefits:

- Reacts with primary amines to add protected sulfhydryls
- When a free sulfhydryl is needed, an easy deprotection step is performed, generating a thioacetylated peptide; this free sulfhydryl-containing peptide can then be conjugated to form a hapten-carrier conjugate

Sulfo-NHS Acetate

A water-soluble acylating reagent that blocks primary amines.



Sulfo-NHS Acetate
M.W. 259.17

Features/Benefits:

- Reacts mainly with primary amines (lysine epsilon amino groups) at pH 7 or greater
- Irreversible reaction is accompanied by the release of *N*-hydroxysulfosuccinimide

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
26777ZZ	Sulfo-NHS Acetate (Sulfosuccinimidyl Acetate)	100 mg	\$ 85

Triethylamine

Can be used in a weak anion exchange HPLC method as an alternative to resolving tryptic peptides on a reverse-phase column.

Features/Benefits:

- Clear, colorless liquid distilled in glass
- Typical purity is 99.5% by GC; less than 0.2% water by Karl Fischer
- Combined use of weak anion exchange and reverse-phase HPLC methods can provide excellent improvement in resolving power for a number of peptide separations
- Angiotensins, diastereomers and analogs of neurotensin and underivatized dipeptides are reported to be resolved with mixtures of triethylammonium acetate buffer (prepared by titrating a 0.01 M acetic acid solution with triethylamine to pH 6), and with acetonitrile as eluent on a weak anion exchange column

References

1. Dizdaroglu, M. and Krutzsch, H.C. (1983). *J. Chrom.* **264**, 223-229.
2. Dizdaroglu, M., et al. (1982). *Anal. Biochem.* **123**, 190-193.
3. Dizdaroglu, M., et al. (1982). *J. Chrom.* **245**, 158-162.
4. Dizdaroglu, M. and Simic, M.G. (1980). *J. Chrom.* **195**, 119-126.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
25108ZZ	Triethylamine Purity: ≥ 99.5% Density: 0.728 Boiling Point: 89°C	100 gm	\$ 42

Special Product

TFA (Trifluoroacetic Acid)

All trifluoroacetic acids are not equal – demand what is crucial for your application.

• Purity

Clear, colorless Pierce Trifluoroacetic Acid (TFA) exhibits superior purity (99.8%) and exceptional clarity, allowing for sensitive, nondestructive peptide detection at low UV wavelengths in reverse-phase HPLC protein and peptide separation systems.¹

• Versatility

Pierce TFA also performs incomparably in protein sequencing applications^{2,3} and solid-phase peptide synthesis⁴ and as a protein/peptide solubilizing agent.^{2,3}

• High-performance Packaging

TFA is an extremely corrosive organic solvent that has vapors that corrode conventional bottle caps. Damaged packaging endangers TFA purity and it sabotages one's work, wasting valuable time. Because packaging is as important as product quality, Pierce packages its TFA in amber glass with protective Teflon® TFE-lined fluorocarbon caps or ampuled under nitrogen.

• Economical Convenience

- Pierce TFA is available in a variety of package formats and sizes, allowing researchers to save money by choosing the package that works best for their specific applications. For example, the 1 ml ampules provide a simple way to prepare liter quantities of 0.1% TFA for stationary and mobile phases in reverse-phase chromatography.

References

- Chicz, R.M. and Regnier, F.E. (1990). High-Performance Liquid Chromatography Effective Protein Purification by Various Chromatographic Methods, In M.P. Deutscher (Ed.), *Guide to Protein Purification, Methods in Enzymology* (pp. 392-421). Academic Press.
- Smith, B.J. (1997). *Protein Sequencing Protocols*. Humana Press. (Product # 20016ZZ)
- Allen, G. (1989). *Sequencing of Proteins and Peptides, Second Revised Edition*. Elsevier.
- Stuart, J.M. and Young, J.D. (1984). *Solid Phase Peptide Synthesis, Second Edition*. Pierce Chemical Company.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
53102ZZ	Trifluoroacetic Acid, HPLC Grade	10 x 1 ml	\$ 7
28901ZZ	Trifluoroacetic Acid, Sequanal Grade	500 ml	\$22
28902ZZ	Trifluoroacetic Acid, Sequanal Grade	10 x 1 gm	\$ 5
28903ZZ	Trifluoroacetic Acid, Sequanal Grade	100 gm	\$ 9
28904ZZ	Trifluoroacetic Acid, Sequanal Grade	10 x 1 ml	\$ 5

Urea

A low UV-absorbing protein denaturant.

Features/Benefits:

- Melting point: 132-136°C
- Specification: $A_{280} < 0.100$

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
29700ZZ	Urea, Sequanal Grade	1 kg	\$ 2

3

Internet: <http://www.pierce.com>

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Enzyme Cleavage Products

This section describes the Pierce line of Proteases for use in protein cleavage applications.

Guide to Pierce Proteases for Protein Analysis

Product #	Available Enzyme	Available Immobilized	Soluble	Specificity
20238ZZ, 20214ZZ	Aminopeptidase M	X	X	• Cleaves amino acids sequentially from the amino terminus
20236ZZ, 20212ZZ	Carboxypeptidase Y	X	X	• Cleaves amino acids sequentially from the carboxy terminus
32520ZZ, 32521ZZ	Factor Xa		X	• Cleaves after the carboxyl side of the Ile-Glu-Gly-Arg sequence
20341ZZ	Papain	X		• Nonspecific protein digestion
20343ZZ	Pepsin	X		• Nonspecific protein digestion
20195ZZ	<i>Staphylococcus aureus</i> Protease	X	X	• Cleaves at the carboxyl side of Aspartic and Glutamic Acid
20199ZZ	Submaxillaris Protease		X	• Cleaves at the carboxyl side of Arginine
20230ZZ, 20233ZZ	TPCK Trypsin	X	X	• Cleaves on the carboxyl side of Arginine and Lysine
20151ZZ	V-8 Protease	X		• Cleaves on the carboxyl side of Glutamic and Aspartic Acids

Aminopeptidase M and Immobilized Aminopeptidase M

Ideal for removing amino acids sequentially from the NH₂-terminus.

Feature/Benefit:

- One unit of enzyme activity hydrolyzes one μ mole of L-leucine-4-nitroanilide at 25°C and pH 7

Reference

1. Royer, G.P. and Andrews, J.P. (1973). *J. Biol. Chem.* **248**, 1807-1812.

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
20214ZZ	Aminopeptidase M (Hog kidney microsomes) Specific Activity: 4 units/mg Supplied: 3.2 M (NH ₄) ₂ SO ₄ , pH 6.9; 10 mM MgCl ₂ Storage: 5°C	20 units	\$129
20238ZZ	Immobilized Aminopeptidase M Supplied: 50% glycerol, 0.02% NaN ₃ Support: Cross-linked 4% beaded agarose Storage: 5°C	1 unit (2 ml gel)	\$ 64

Carboxypeptidase Y and Immobilized Carboxypeptidase Y

Effectively remove amino acids sequentially from the COOH terminus.

Reference

1. Martin, B., et al. (1971). *Carlsberg Res. Comm.* **42**, 99-102.

Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
20212ZZ	Carboxypeptidase Y (Baker's yeast) Specific Activity: 60 units/mg (one unit of enzyme activity hydrolyzes one μ mole of ATEE at 25°C and pH 8) Supplied: Lyophilized in 0.1 M Sodium Citrate, pH 5.3 Storage: 0°C	30 units (0.5 mg)	\$ 62
20236ZZ	Immobilized Carboxypeptidase Y Supplied: 50% glycerol, 0.02% NaN ₃ Support: Cross-linked 4% beaded agarose Spacer: Diaminodipropylamine Storage: -20°C	25 units (2 ml gel)	\$ 88

800-874-3723

ConnectaSH™ Protein Disulfide-Isomerase

Accelerates the exchange reactions between disulfide bonds in proteins in the presence of an appropriate oxidizing or reducing agent ... it unscrambles proteins.

References

1. Goldberger, R.F., et al. (1964). *J. Biol. Chem.* 239, 1406.
2. Lambert, N. and Freedman, R.B. (1983). *Biochemistry J.* 213, 225.
3. Hillson, D.A., et al. (1984). *Meth. in Enzymology* 107, 281.
4. Tang, J.G., et al. (1988). *Biochemistry J.* 255, 451.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20197ZZ	ConnectaSH™ Protein Disulfide-Isomerase*	1 mg	\$378
Activity: Approximately 40% of the activity of scrambled RNase is recovered at room temperature in 60 minutes by adding 20 µg/ml Protein Disulfide-Isomerase			

*This item requires special shipping on dry ice at an additional charge.

Factor Xa

A restriction endoprotease that releases peptides from fusion proteins.

Features/Benefits:

- Extremely specific proteolytic activity – useful in the genetic engineering of proteins
- Cleaves after the last amino acid in the sequence Ile-Glu-Gly-Arg
- Activity is > 130 U/mg using a synthetic Ile-Glu-Gly-Arg peptide with *p*-nitroanilide derivatized arginine as the substrate; each unit of enzyme cleaves 1 µmole/minute of substrate at 37°C in 5 mM MES, 0.5 M NaCl, 1 mM benzamidate at pH 6 with 1% BSA

References

1. Nagai, K. and Thøgersen, H. (1984). *Nature* 309, 810-812.
2. Aurell, L., et al. (1984). *Thrombosis Res.* 11, 595-609.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
32521ZZ	Factor Xa (Bovine)*	50 µg	\$ 59
32520ZZ	Factor Xa (Bovine)* Supplied in 5 mM MES, pH 6, 0.5 M NaCl Protein Concentration: 1-2 mg/ml	250 µg	\$236

*This item requires special shipping on dry ice at an additional charge.

Papain (Immobilized) and Pepsin (Immobilized)

Produce active immunoglobulin fragments.

Features/Benefits:

- Pepsin and papain can cleave Fc fragments from IgG, resulting in F(ab')₂ or Fab fragments, respectively
- These enzymes have a broad specificity, including esterase activity
- Immobilized enzymes result in easier enzyme treatments; to stop the enzymatic digestion, simply remove the agarose beads

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20341ZZ	Immobilized Papain Support: Cross-linked 6% beaded agarose Activity: 7 BAEE units per ml of settled gel Loading: 250 µg per ml of gel	5 ml gel	\$125
20343ZZ	Immobilized Pepsin Support: Cross-linked 6% beaded agarose Activity: 2,000-4,000 units per ml of settled gel Loading: 2-3 mg per ml of gel	5 ml gel	\$125

3

Immobilized Pepsin/Immobilized Papain

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QuantiCleave™ Protease Assay Kit

Detects protease levels as low as 1 ng/ml!

Features/Benefits:

- Three times faster and 1,000 times more sensitive than unmodified casein-based protease assays
- Requires just half the sample of unmodified casein-based protease assays
- No corrosive precipitants
- Entire assay can be run in a microwell plate
- Easy-to-perform – with results in less than one hour
- Simultaneous measurement of multiple samples in an ELISA plate reader
- Manipulation of time, temperature and pH allows optimization of sensitivity

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
23263ZZ	QuantiCleave™ Protease Assay Kit <i>Sufficient for 250 assays.</i>	Kit	\$189
	Includes: Succinylated Casein	5 x 10 mg	
	TNBSA (5% w/v aqueous)	2 ml	
	Trypsin Standard (40 BAEE units/mg)	50 mg	
	BupH™ Borate Buffer Pack (yields 500 ml)	1 pack	

Staphylococcus aureus Protease

Cleaves specifically at the carboxy side of aspartic and glutamic acids.

Features/Benefits:

- Specificity for glutamic acid is achieved in ammonium bicarbonate, pH 7.8 or ammonium acetate, pH 4
- Enzyme is also active in the presence of many denaturing agents such as SDS, Urea and Guanidine•HCl

References

1. Drapeau, G.R., et al. (1972). *J. Biol. Chem.* 247, 6720.
2. Houmard, J. and Drapeau, G.R. (1972). *PNAS* 69, 3506.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20195ZZ	<i>Staphylococcus aureus</i> Protease Source: V-8 strain Activity: 500-700 units/mg (with casein as the substrate) Supplied: Lyophilized and salt-free Storage: Dessicated, below 0°C	5 mg	\$225

Submaxillaris Protease

Cleaves specifically at the -COOH side of arginine.

Reference

1. Schenkein, et al. (1977). *Arch. Biochem. Biophys.* 182, 64.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20199ZZ	Submaxillaris Protease Source: Mouse submaxillary gland Specific Activity: 250-300 units/mg Supplied: Lyophilized and salt-free Storage: 0°C	250 units (1 mg)	\$231

TPCK Trypsin and Immobilized TPCK Trypsin

Cleave specifically at the -COOH side of arginine and lysine.

Feature/Benefit:

- Immobilized TPCK Trypsin makes it easier to eliminate trypsin contamination in tryptic digests

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20230ZZ	Immobilized Trypsin, TPCK Treated Source: Bovine pancreas Activity: ≥ 200 TAME units/ml Supplied: 50% glycerol, 0.05% NaN ₃ Support: Cross-linked 4% beaded agarose Storage: 5°C	2 ml gel	\$ 56
20233ZZ	TPCK Trypsin (2,000 BAEE Units)	50 mg	\$ 58

800-874-3723

V-8 Protease, Immobilized

Unique coupling method ensures that the immobilized protease does not autolyse.

Features/Benefits:

- Eliminates protease contamination in samples
- No quenching required
- Stable in SDS, Urea and Guanidine
- Three V-8 Digestion Buffers included are specific for glutamic acid residues or glutamic and aspartic acid residues

References

1. Regnier, F. (1987). *LC-GC* 5(5), 392-398.
2. Jirgensons, B. (1981). *Biochimica et Biophysica Acta* 699, 206-209.
3. Drapeau, G. (1978). *J. Biol. Chem.* 253(17), 5899-5901.
4. Drapeau, G. (1978). *Canadian J. Biochem.* 56(6), 534-544.
5. Drapeau, G., et al. (1972). *J. Biol. Chem.* 247(20), 6720-6726.
6. Houmard, J. and Drapeau, G. (1972). *Proc. Natl. Acad. Sci. USA* 69(12), 3506-3509.

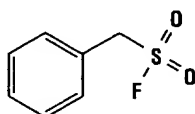
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20151ZZ	Immobilized V-8 Protease Kit <i>Contains enough enzyme and buffers to perform up to 20 digestions of 2 mg protein samples.</i>	Kit	\$498
	Includes: Immobilized <i>S. aureus</i> Protease (Activity: 35-60 units/ml of gel with casein as substrate)	2.5 ml	
	V-8 Digestion Buffer 1	200 ml	
	V-8 Digestion Buffer 2	200 ml	
	V-8 Digestion Buffer 3	200 ml	
	Accessories (used to separate agarose)		

Inhibitors (Proteolytic Enzymes)

PMSF

Reacts with serine residues to inhibit trypsin, chymotrypsin, thrombin and papain.



PMSF
M.W. 174.19

Features/Benefits:

- High salt concentrations are used to reverse the binding (and inhibition) of PMSF to enzymes
- Because PMSF has limited water solubility, it must be dissolved in a small amount of solvent, ethanol or methanol before addition to a buffer

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
36978ZZ	PMSF (Phenylmethylsulfonyl Fluoride)	5 gm	\$ 4

Compatible Products

51102ZZ	Ethanol (HPLC Grade)	1 liter	\$ 3
54124ZZ	Methanol (HPLC Grade)	1 liter	\$ 2

Soybean Trypsin Inhibitor, Immobilized

For effective removal of trypsin, chymotrypsin and elastase from protein digests.

Applications:

- Purifying trypsin, chymotrypsin and elastase^{1,2}
- Removing proteases from activated pancreatic juices³

References

1. Feinstein, G., et al. (1974). *Euro. J. Biochem.* 43, 569-581.
2. Peterson, L.M., et al. (1976). *Biochemistry* 15, 2501-2508.
3. Reek, G.R., et al. (1971). *Biochemistry* 10, 4690-4698.

Ordering Information

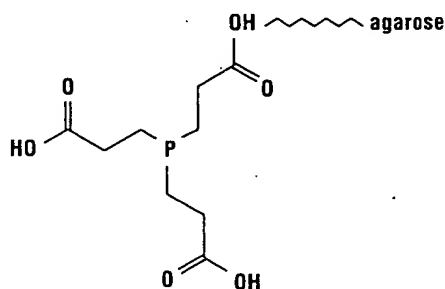
Product #	Description	Pkg. Size	U.S. Price
20235ZZ	Immobilized Soybean Trypsin Inhibitor <i>STI coupled to spherical 4% beaded agarose.</i> Capacity: Minimum of 9 mg trypsin per ml of gel Supplied: Suspension of 50% glycerol containing 0.05% Na ₂ S ₂ O ₅	2 ml gel	\$ 7

Reducing Agents

Tris(2-carboxyethyl)phosphine, commonly known as TCEP, is an excellent disulfide reducing agent that yields selective and complete reduction of even the most stable water-soluble alkyl disulfides.

Bond-Breaker™ TCEP Disulfide Reducing Gel

An extremely efficient, versatile and odorless reducing agent.



Immobilized TCEP

Features/Benefits:

- Versatile, solid-phase, odorless reduction of disulfide bonds
- Outstanding reducing activity, stability and efficiency at an economical price
- Ready-to-use – no pretreatment or refrigeration required

- Effectively reduces peptides and proteins at room temperature (peptides: instantaneously; proteins: 5-60 minutes)
- Immobilized gel format eliminates the removal of contaminating soluble reductants
- Because it's nonsulfhydryl-based, there's no interference with maleimide chemistry (and other) applications due to interfering free-sulfhydryls
- Available in convenient formats: 5 x 1 ml columns or 5 ml bulk pack

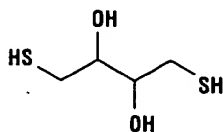
Ordering Information:

Product #	Description	Pkg. Size	U.S. Price
77710ZZ	Bond-Breaker™ TCEP Disulfide Reducing Gel (Tris[2-carboxyethyl]phosphine on 6% cross-linked agarose)	5 x 1 ml pre-packed columns	Please inquire
77711ZZ	Bond-Breaker™ TCEP Disulfide Reducing Gel	5 ml	Please inquire

800-874-3723

DTT

A water-soluble reagent that reduces disulfide bonds.



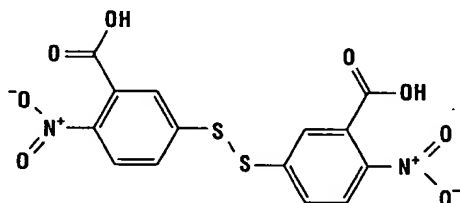
DTT
M.W. 154.25

Applications:

- Maintain mono-thiols completely in the reduced state and reduce disulfide bonds quantitatively
- Specific and sensitive assay for disulfides using DTT with Ellman's Reagent

Ellman's Reagent

A simple, sensitive and reliable method for determining free sulfhydryl content in peptides, proteins and tissues.



Ellman's Reagent
M.W. 396.35

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20290ZZ	DTT, Cleland's Reagent (Dithiothreitol)	5 gm	\$ 73

Compatible Product

22582ZZ	Ellman's Reagent	5 gm	\$ 45
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Reference

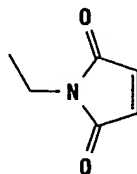
1. Ellman, G.L. (1959). *Arch. Biochem. Biophys.* 82, 70-77.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
22582ZZ	Ellman's Reagent (5,5'-Dithio-bis-(2-nitrobenzoic Acid))	5 gm	\$ 45

N-Ethylmaleimide

Useful for blocking free sulfhydryls after reduction.



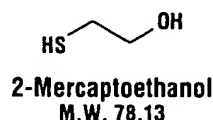
N-Ethylmaleimide
M.W. 125.13

Features/Benefits:

- Forms covalent bonds with sulfhydryl groups at pH 6.5-7.5
- Reaction can be monitored by measuring the decrease in absorbance at 300 nm
- Has been used successfully for the blocking of sulfhydryl-containing reagents that interfere in enzyme assays

Ordering Information

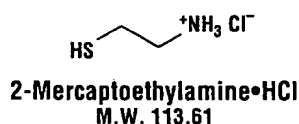
Product #	Description	Pkg. Size	U.S. Price
23030ZZ	N-Ethylmaleimide (NEM)	25 gm	\$ 85

2-Mercaptoethanol*A mild reducing agent for cleaving disulfide bonds to thiols.***Features/Benefits:**

- Also known as β -Mercaptoethanol
- Often included in enzyme solutions to protect against catalytic site inactivation due to cysteine sulphydryl oxidation/disulfide formation; added at final concentrations of 5 and 20 mM, with or without EDTA as an additional protectant

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
35601ZZ	2-Mercaptoethanol	6 x 1 ml	\$ 27
35600ZZ	2-Mercaptoethanol	500 gm	\$ 33

2-Mercaptoethylamine•HCl*Dissociates divalent IgG to monovalent IgG without dissociating heavy and light chains.***Ordering Information**

Product #	Description	Pkg. Size	U.S. Price
20408ZZ	2-Mercaptoethylamine•HCl	6 x 6 mg	\$ 92

Reduce-Imm™ Reducing Kit*Solid-phase system for reducing peptides and proteins.*

Both the mercaptan and the DTT methods of reduction use soluble reducing agents requiring that the reducing reagents be removed from the sample before further procedures can be performed. The Reduce-Imm™ Reducing Kit was developed to eliminate this requirement and is highly effective for reducing disulfide bonds.

Features/Benefits:

- Eliminates soluble components that interfere with the assay of free thiol groups
- No need to remove reducing agent after the reaction
- Reduced protein, free of contaminating thiols, can be easily assayed using the BCA method
- Excellent protein recovery
- Regenerable matrix useable up to four times

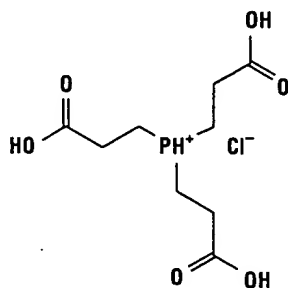
Ordering Information

Product #	Description	Pkg. Size	U.S. Price
77700ZZ	Reduce-Imm™ Reducing Kit Sufficient reagents to reduce 150 μ moles of disulfide. Includes: Reduce-Imm™ Immobilized Reductant Columns (capacity: 30-40 μ moles of sulphydryl groups per column) Reduce-Imm™ Equilibration Buffer #1 Reduce-Imm™ Equilibration Buffer #2 DTT (used to regenerate and prime the column) Ellman's Reagent	Kit 2 x 2 ml 500 ml 250 ml 250 mg 500 mg	\$299
77701ZZ	Reduce-Imm™ Immobilized Reductant Column	1 x 2 ml	\$109

• 800-874-3723

TCEP HCl

Potent, water-soluble, odorless reducing agent in a conventional format.



TCEP·HCl
M.W. 286.65

Features/Benefits:

- Selective and complete reduction of even the most stable water-soluble alkyl disulfides

- Effective reduction at room temperature and pH 5 in less than five minutes
- Water solubility of 310 gm/L
- Resistant to air oxidation; nonvolatile and nonreactive toward other functional groups found in proteins

Reference

1. Kirley, T.L. (1989). *Anal. Biochem.* 180, 231.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20490ZZ	TCEP·HCl (Tris[2-carboxyethyl]phosphine hydrochloride)	1 gm	\$ 34

Renaturants and Preservatives

ConnectaSH™ Protein Disulfide-Isomerase

Catalyzes protein disulfide formation – renatures reduced or scrambled proteins.

Features/Benefits:

- Accelerates the exchange reactions between disulfide bonds in proteins in the presence of an appropriate oxidizing or reducing agent
- Isolated from bovine liver and has a molecular weight of 107,000
- Approximately 40% of the activity of scrambled RNase is recovered at room temperature in 60 minutes by adding 20 µg/ml Protein Disulfide-Isomerase (PDI)
- Scrambled ribonuclease (ribonuclease that has been reduced and then reoxidized under denaturing conditions) can be used to assay PDI activity; in the presence of PDI, ribonuclease activity is recovered from scrambled, inactive ribonuclease

Applications:

- Reforming native insulin from scrambled insulin or from the separate S-sulfonated insulin chains with 20-30% yields⁴
- Use in the presence of 1 M guanidine hydrochloride to facilitate the refolding of human pro-urokinase that had been expressed in *Escherichia coli*⁵

- Partially purified PDI from rat liver used to recover 18% of the activity from reduced lysozyme over a two minute incubation¹
- PDI isolated from mouse used to renature reduced IgM with yields up to 50%⁶
- Renaturing bovine serum albumin, soybean trypsin inhibitor and bovine pancreatic trypsin inhibitor^{3,7}

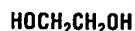
References

1. Goldberger, R.F., et al. (1964). *J. Biol. Chem.* 239, 1406.
2. Lambert, N., et al. (1983). *Biochemistry J.* 213, 225.
3. Hillson, D.A., et al. (1984). *Meth. in Enzymol.* 107, 281.
4. Tang, J.G., et al. (1988). *Biochemistry J.* 255, 451.
5. Okumura, K., et al. (1988). *Agric. Biol. Chem.* 52, 1735-1739.
6. Koshland, M.E., et al. (1981). *Biochemistry* 20, 6594-6599.
7. Freedman, R.B., et al. (1980). *The Enzymology of Post-Translational Modifications of Proteins*. Vol. I. New York: Academic Press, p.157.

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
20197ZZ	ConnectaSH™ Protein Disulfide-Isomerase*	1 mg	\$378

*This item requires special shipping on dry ice at an additional charge.

Ethylene Glycol*Specially purified to protect your proteins.***Ethylene Glycol**
M.W. 62.07**Features/Benefits:**

- Specially purified to remove impurities such as aldehydes, peroxides, iron and UV-absorbing hydrocarbons
- Suitable for enzyme storage without the worry of losing enzymatic activity
- Stable for months

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
29810ZZ	Ethylene Glycol (50% aqueous solution)	200 ml	\$ 71

FreeZyme® Conjugate Purification Kit*Improves the sensitivity of antibody-enzyme conjugates by removing excess free enzyme.***Features/Benefits:**

- Reduced background for blotting and ELISA
- Fewer washes needed to remove background
- Increased sensitivity with more active reagent
- Greatly improved signal-to-noise ratio

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
44920ZZ	FreeZyme® Conjugate Purification Kit* Sufficient materials for 10 purifications of 0.5-50 mg of antibody-enzyme conjugate; kit is designed for both horseradish peroxidase and alkaline phosphatase conjugates.	Kit	\$320
	Includes: Immobilized IDA	1 x 2 ml	
	Desalting Column	1 x 10 ml	
	FreeZyme® Column Activator	25 ml	
	FreeZyme® Elution Buffer	35 ml	
	BupH™ Tris Buffered Saline	2 packs	
	BupH™ Phosphate Buffered Saline	1 pack	
	Alkaline Phosphatase Activity Preserving Solution	1 ml	

*U.S. Patent # 5,266,686

Protein Disulfide-Isomerase

See ConnectaSH™ Protein Disulfide-Isomerase on page 254.

SuperFreeze™ Conjugate Stabilizers*Protect enzyme conjugates during storage.***Features/Benefits:**

- Provide a buffered antifreeze environment for enzyme conjugated antibodies and proteins
- Maintain a liquid, low-viscosity solution at -20°C
- No aliquoting of conjugate is necessary and sampling is convenient
- Contain preservatives to prevent microbial growth during long-term storage

Ordering Information

Product #	Description	Pkg. Size	U.S. Price
31502ZZ	SuperFreeze™ Phosphatase Conjugate Stabilizer	25 ml	\$ 35
31503ZZ	SuperFreeze™ Peroxidase Conjugate Stabilizer	25 ml	\$ 35

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